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Nitrous oxide emissions and mitigation strategies in winter oilseed rape cultivation

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"Imagination is more important than knowledge. For knowledge is limited, whereas imagination embraces the entire world, stimulating progress, giving birth to evolution."

Albert Einstein

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Frequently used abbreviations

ASN	ammonium sulphate nitrate
atom% ^{15}N	^{15}N abundance in atom percentage
CAN	calcium ammonium nitrate
CH_4	methane
CO_2	carbon dioxide
DMPP	3,4-dimethylepyrazole phosphate
ECD	electron capture detector
EF	emission factor
FID	flame ionization detector
IPCC	Intergovernmental Panel on Climate Change
MC	microcosm
N	nitrogen
N_2	dinitrogen
N_2O	nitrous oxide
NH_4	ammonium
N_{min}	mineral nitrogen
NO_3	nitrate
OSR	winter oilseed rape
SE	standard error
TZ+MP	1H-1,2,4-triazole and 3-methylpyrazole
WFPS	water-filled pore space

Abstract

After carbon dioxide (CO_2) and methane (CH_4), nitrous oxide (N_2O), is the third most important greenhouse gas (GHG) in the atmosphere. Nitrous oxide contributes to the greenhouse gas effect as well as to ozone depletion. The major portion of anthropogenic N_2O emissions are stimulated by the use of nitrogen fertilizers in agriculture. The main processes for N_2O production in soils are nitrification and denitrification. Various environmental and management factors such as precipitation, soil type, tillage, and crop residues affect these processes.

N_2O emissions can occur substantially in the post-harvest period. In Germany, approximately 50 % of the annual N_2O emissions can occur during winter. This exhibits the importance and necessity of annual data sets which prevent misinterpretations instigated by investigations limited to the vegetation period.

Winter oilseed rape (*Brassica napus* L.; OSR) is the most important raw material for biodiesel in Germany. As of 2018, the framework of the European Renewable Energy Directive requires that the use of biofuels achieve GHG savings of at least 50 % compared to fossil fuels. Feedstock production for biodiesel contributes more than half of the total GHG emissions. To close the nutrient cycle with renewable energy, digestate from biogas plants can be used as a substitute for mineral N fertilizer permitting the reduction of GHG emissions in the production process of synthetic fertilizers. When compared to other crops, OSR has a high N demand. The low N removal by the seeds results in inefficient use of nitrogen and therefore a high N surplus in the soil which is susceptible to gaseous or leaching losses to the environment. Another potential risk for N_2O losses are crop residues after harvest.

The type of soil cultivation can have both positive and negative implications on N_2O emissions which depend, among other things, on tillage depth, soil type and moisture. Results from studies measuring N_2O emissions from different tillage systems are contradicting and site dependent.

This study aims to investigate the effect of *(a)* N fertilization (mineral and organic), *(b)* nitrification inhibitors, *(c)* crop residues and *(d)* tillage on direct N_2O emissions and, inter alia, yield and soil nitrogen dynamics in OSR production.

N_2O emissions were monitored for three years over a range of N fertilization levels at five study sites chosen so as to best represent typical winter oilseed rape production in Germany. Furthermore, the effect of the nitrification inhibitor (NI) TZ+MP (1H-1,2,4-triazole and 3-methylpyrazole) with digestate is investigated. Additional experiments for ^{15}N labelled crop residues, nitrification inhibitor DMPP (3,4-dimethylepyrazole phosphate) with mineral fertilizer and soil tillage were implemented.

A high spatial and temporal variability in N_2O fluxes over all sites was observed. At each site, increased N_2O fluxes were often detected after N fertilization in conjunction with rainfall events. During the first six weeks after harvest we frequently observed increased N_2O fluxes following rainfall. In this postharvest period of winter oilseed rape, nitrate contents in the top

soil were generally elevated. There were no considerable N_2O pulses observed during thawing of frozen soil. Winters were mild without any severe frost periods in all three surveyed years which could be a reason for the generally low N_2O winter fluxes observed in this study. On all examined sites, increasing N fertilization significantly enhanced N_2O flux rates.

Data obtained during the study were used to augment an existing model, wherefrom a new emission factor for OSR can be calculated. Assuming a quantity of 200 kg N ha^{-1} the global fertilizer-related emission factor derived from the exponential model was 0.6 %. This factor is within the uncertainty range of the EF1 IPCC emission factor (0.3 % – 3.0 %), but about 40 % lower than the 1 % IPCC default.

The nitrification inhibitor (NI) TZ+MP combined with digestate mitigated the N_2O fluxes significantly across all study sites and experimental years. As already noted in the fertilizer experiment, a high spatial and temporal variability in N_2O fluxes over all sites was observed. The magnitudes of the N_2O fluxes also showed similar trends. Over the entire investigation, the application of the NI significantly reduced annual N_2O emission by a factor of three. During the fertilization period this mitigation effect was six times significant. This clearly emphasizes the importance of annual data sets to avoid overestimating NI effects.

In field experiments with crop residues, increased N_2O fluxes were measured immediately after ^{15}N crop residue application in conjunction with precipitation. Between 50 – 68 % of the total N_2O emission occurred during the post-harvest period, highlighting the importance of that period for N_2O budgets in OSR production. However, crop residues or tillage did not affect total cumulative N_2O emission. Only 4.2 % (conventional tillage) and 5.2 % (reduced tillage) of the N released as N_2O during the investigated period stemmed directly from the applied OSR residues. The low contribution from crop residues on total N_2O emissions presumably resulted from the large C/N-ratio (52) of the OSR residues.

The study has indicated that with a moderate N fertilization (120 kg N ha^{-1}) GHG saving goals can be reached with the same amount of OSR oil yield compared to higher N fertilization rates. In addition, NIs are an effective tool to mitigate N_2O emissions from OSR, depending on site and year.

Zusammenfassung

Lachgas (N_2O) ist nach Kohlendioxid (CO_2) und Methan (CH_4) das drittwichtigste klimawirksame Spurengas in der Atmosphäre. N_2O ist am Treibhauseffekt, sowie am Ozonabbau beteiligt. Der größte Anteil der anthropogenen N_2O-Emissionen wird durch die Landwirtschaft verursacht, hier hauptsächlich durch den Einsatz von Stickstoffdüngern. Stickstoff (N) in unterschiedlichen Verbindungen, ist das Ausgangssubstrat aller N_2O-bildenden Prozesse in Böden. Die wichtigsten Prozesse sind hierbei die Nitrifikation und die Denitrifikation. Verschiedene Steuergrößen wie beispielsweise die Niederschlagsintensität, Bodenart, Bodenbearbeitung, sowie Ernterückstände, beeinflussen die Größenordnung der gebildeten N_2O-Emissionen. Die Bewirtschaftung beeinflusst jedoch nicht nur die Bildung von N_2O während der Vegetationsperiode, sondern hat auch Auswirkungen auf die drauffolgenden Nachernteemissionen. In Deutschland können Winteremissionen bis zu 50 % der gesamten N_2O-Jahresemission betragen. Dies macht deutlich, wie bedeutend ganzjährige Messkampagnen für eine aussagekräftige Klimawirksamkeit von Anbausystemen sind.

Winterraps (*Brassica napus* L.) ist der wichtigste Rohstoff für die Biodieselproduktion in Deutschland. Die im Jahr 2009 von der Europäische Union verabschiedete Richtlinie für erneuerbare Energie (Renewable Energy Directive; *RED*, 2009), zählt zu den wichtigsten Nachhaltigkeitsbestimmungen rund um die Produktion von Biokraftstoffen. In dieser Richtlinie erfolgte unter anderem die Festlegung, dass Biodiesel ab 2018 gegenüber fossilen Brennstoffen ein Treibhausgasminderungspotential von mind. 50 % aufweisen muss. Mehr als die Hälfte der Treibhausgase in der Biodieselherstellung entstehen während der Rohstoffproduktion in der Landwirtschaft, N_2O kann bis zu 88 % daran beteiligt sein (*Hoefnagels* et al., 2010). Winterraps ist im Vergleich zu anderen Kulturpflanzenarten durch einen hohen Stickstoffbedarf charakterisiert. Durch eine ungünstige N–Ausnutzung besteht überdies ein besonderes Risiko von Umweltbelastungen durch N–Austräge in die Umwelt. In der Literatur werden von Stickstoffüberschüssen bis zu 90 kg N ha^{-1} berichtet, welche ein hohes Potential für N_2O–Emissionen, sowie Nitratauswaschung darstellen. Eine weitere potenzielle Quelle für Lachgasemissionen bilden die auf dem Feld zurück belassenen Ernterückstände.

In dieser Studie wird der Einfluss der Stickstoffdüngung (mineralisch und organisch), Nitrifikationsinhibitoren (zu mineralischem und organischem N-Dünger), Ernterückständen und Bodenbearbeitung auf direkte N_2O-Emissionen, Ertrag und der Bodenstickstoffdynamik im Rapsanbau untersucht. Über drei Jahre hinweg wurden in mindestens wöchentlichen Abständen an fünf verschiedenen Standorten in Deutschland N_2O-Flussraten in Winterraps, so wie in den Folgekulturen Winterweizen und Wintergerste, gemessen. An den jeweiligen Standorten wurde ein Parzellenversuch mit einheitlichen Versuchsvarianten angelegt, an welchen der Einfluss von unterschiedlicher Stickstoffmenge als Mineraldünger auf annuelle N_2O–Emissionen untersucht wurde. Zusätzlich wurden Untersuchungsvarianten zur organischen Düngung (Gärsubstrat) mit und ohne Nitrifikationshemmstoff (NI) angelegt. Am

Standort Hohenheim (Versuchsstation Ihinger Hof) wurden ergänzend Versuche mit Ernterückständen mittels ^{15}N-Isotopentracer durchgeführt. Zudem wurde der Einfluss eines NI zur mineralischen Düngung und der Einfluss der Bodenbearbeitung auf N_2O–Emissionen untersucht.

Die N_2O–Flussraten der Standorte zeigten über die Studie hinweg eine hohe räumliche und zeitliche Variabilität. Erhöhte Flüsse wurden häufig nach der Düngung in Zusammenhang mit Niederschlagsereignissen gemessen. Auch in den ersten sechs Wochen der Nacherntеperiode wurden vielfach erhöhte N_2O-Raten nach Niederschlag gemessen. Während dieses Zeitraums waren die Nitratgehalte des Oberbodens allgemein erhöht. Innerhalb der Projektlaufzeit wurden an keinem Standort nennenswerte Frost-Tau-induzierte N_2O–Emissionen beobachtet. Allgemein waren die gemessenen N_2O–Flussraten der gesamten Studie niedrig. Über alle Standorte hinweg konnte ein Effekt der Düngung nachgewiesen werden. Am Standort Merbitz (toniger Boden) wurden die höchsten, an den sandigen Standorten Berge und Dedelow die niedrigsten Flussraten gemessen. Eine Ausnahme stellte der Standort Hohenschulen, welcher trotz hohem Sandanteil, deutlich höhere N_2O–Emissionen aufwies als die übrigen sandigen Standorte. Die erhobenen düngerinduzierten N_2O-Emissionen wurden in ein bestehendes Modell (*Walter* et al. 2015) implementiert, wodurch ein neuer Emissionsfaktor für Winterraps berechnet werden konnte. Bei einer Düngermenge von 200 kg N ha^{-1} ermittelte das Exponentialmodell einen düngerbezogenen Emissionsfaktor von 0,6 %. Dieser Wert liegt innerhalb des Unsicherheitsbereichs (0,3 % – 3,0 %) des IPCC Emissionsfaktor EF1, liegt jedoch 40 % niedriger als der vom IPCC ausgegebene Standardwert von 1 %.

Der Nitrifikationshemmstoff zeigte über alle Jahre und Standorte hinweg eine signifikante Minderung der N_2O-Emissionen. Eine signifikante Minderung der annuellen N_2O Emissionen aufgrund der NI Anwendung konnte lediglich dreimal während des untersuchten Zeitraums gemessen werden. Während der Düngeperiode waren die Emissionen gegenüber den Kontrollen ohne NI in sechs Fällen signifikant reduziert. Dieses Ergebnis unterstreicht die Bedeutung der annuell gemessenen N_2O Emissionen, wodurch einer Überschätzung der Hemmstoffwirkung von NIs entgegengewirkt werden kann. Die Anwendung von NI am Ihinger Hof bei mineralischer Düngung führte nur im ersten Versuchsjahr zu der erwarteten Reduktion der N_2O-Emissionen (17 %).

Je nach Bodenbearbeitung verursachten die ^{15}N-makierten Ernterückstände einen Anteil zwischen 4,2 (Pflug) und 5,2 (reduzierte Bodenbearbeitung) % an der annuellen N_2O-Emission. Zurückzuführen war dies auf den hohen C/N-Wert der Ernterückstände (52). In diesem Versuch wurden 50–68 % der gesamten N_2O–Emissionen in der Nacherntеperiode emittiert. Über den gesamten Messzeitraum hinweg hatten jedoch weder die Ernterückstände, noch die unterschiedliche Bodenbearbeitung einen signifikanten Einfluss auf die kumulativen N_2O-Emissionen. Unterstrichen wurden diese Ergebnisse durch einen Mikrokosmenversuch mit Ernterückständen unterschiedlicher C/N–Qualität und Mengen. Auch hier wurden keine Unterschiede zwischen den kumulativen N_2O-Emissionen gemessen. Tendenziell hatte die

unbehandelte Kontrolle die höchsten Emissionen. Von Rapsernterückständen ist somit keine signifikante Erhöhung der N_2O-Emissionen zu erwarten. Auf Grund der hohen C/N–Verhältnisse, kann es vielmehr zu einer Immobilisierung des verfügbaren N, resultierend in einer Hemmung der N_2O Bildung kommen. Erhöhte N_2O-Emissionen in der Nacherntеperiode von Raps sind daher auf den hohen Stickstoffüberschuss, auf Grund der schlechten Stickstoffnutzungseffizienz von Raps zurückzuführen.

Die Ergebnisse der Studie haben aufgezeigt, dass auch mit einer moderaten N–Düngung von 120 kg ha^{-1} keine Einbußen im Rapsölertrag zu erwarten sind, jedoch N–Austräge in die Umwelt reduziert, und das gewünschte Treibhausgas–minderungspotential erreicht werden kann. Darüber hinaus konnte gezeigt werden, dass abhängig von Jahr und Standort, ein Nitrifikationshemmstoff effektiv N_2O– Emissionen von Gärsubstrat reduzieren kann.

1 General introduction

Winter oilseed rape

Winter oilseed rape (*Brassica napus* L., OSR) is the major oil crop in Europe. The production increased from 1993 to 2014 by 43.1 % (*FAOSTAT*, 2017). Rapeseed comprised 70 % of European oilseed production in 2012 (*Carré & Pouzet*, 2014). With 6.7 x 10^6 ha in 2014, OSR covered approximately 4 % of total utilised agricultural land area in EU-28 (*FAOSTAT*, 2017; *EUROSTAT*, 2017). The corresponding mean grain yield was 3.2 Mg ha^{-1}. In 2015, the principal producers in the EU were France with 5.2 x 10^6 t and Germany with 5.0 x 10^6 t, followed by Poland and Great Britain (*STATISTA*, 2017).

Rising demand for OSR was mainly a result of high consumption for biodiesel production, a consequence of the Renewable Energy Directive (*RED*, 2009), which aimed for a biofuel share of 10 % in transport energy demand. OSR oil is the most common feedstock for biodiesel, which is the major biofuel in Europe (*Hamelinck* et al., 2012). Also in Germany OSR is mainly used for biodiesel production. Beside used cooking oil (22 %), OSR covered 70 % of biodiesel feedstock material. Palm and soybean oil play a minor role in Germany (*FNR*, 2017).

In addition, the *RED* (2009) requires from 2018 a saving of associated greenhouse gas (GHG) emissions from biofuel by 50 % in comparison to fossil fuel. Feedstock production for biodiesel contributes between 74 and 90 % of total GHG emissions, whereas nitrous oxide (N_2O) contributes between 44 and 88 % (*Hoefnagels* et al., 2010).

Winter oilseed rape is known for a high N demand during its early growth stages. However, low N removal by harvested seeds results in low nitrogen use efficiency and high N surpluses, the latter being susceptible to gaseous or leaching losses into the environment (*Rathke* et al., 2006). Maximum yields are often achieved with N rates exceeding 200 kg N ha^{-1} whereas N harvest index is low, thereby resulting in high N surpluses of up to 90 kg N ha^{-1} a^{-1} (*Henke* et al., 2007; *Sieling & Kage*, 2010). The challenges of OSR production as well as the environmental impacts are described in more detail in Chapter 4.

In the last decade, the atmospheric N_2O concentration increased by 0.73 ppb a^{-1}. The mean concentration in 2015 was 328 ppb, about 21 % higher than in the pre-industrial period (*WMO*, 2016). N_2O contributes 7.4 % (0.17 W m^{-2}) to the total anthropogenic radiative forcing (*IPCC*, 2013) and participates in ozone layer depletion (*Crutzen*, 1981). N_2O has a 265 times greater global warming potential than carbon dioxide in a 100-yr time horizon (*Mhyre* et al., 2013). Approximately 40 % of total N_2O emissions are anthropogenic. Agricultural soils provide the main part with 4.1 Tg N yr^{-1} (approx. 60 % of total anthropogenic emissions) (Table 1.1). The stratospheric sink minus cumulate sources gives a growth rate of 3.61 Tg N yr^{-1}. Related to the total radiative forcing (7.4 %) mitigation of the OSR production emissions (N_2O) seems negligible, but with regard to the demanded

mitigation of the GHG emissions caused by the feedstock production for biodiesel (50 %) – searching for mitigation strategies in OSR production gain in importance.
In the last decades, two microbial processes in soil were mentioned as the major sources of N_2O production: nitrification and denitrification (*Bremner*, 1997). But apart from these two processes, the share of further microbial and chemical N transformations such as nitrifier denitrification or chemical denitrification to the N_2O release from agricultural soils are currently discussed (*Shaw* et al., 2006; *Butterbach-Bahl* et al., 2013). Today there are four microbial processes known, depending on definition. Denitrification and nitrate ammonification as nitrate or nitrite reducing processes as well as ammonia oxidation (nitrification) and nitrifier denitrification. The processes are shown in Figure 1.1.

Tab. 1.1 : N_2O sources of 2006/2011 (*Clais* et al., 2013).

Anthropogenic sources	**N_2O** Tg N yr^{-1}	**range**
Fossil fuel combustion & industrial processes	0.7	0.2 – 1.8
Agriculture	4.1	1.7 – 4.8
Biomass and biofuel burning	0.7	0.2 – 1.0
Human excreta	0.2	0.1 – 0.3
Rivers, coastal zones	0.6	0.1 – 2.9
Atmospheric deposition	0.6	0.1 – 0.9
Surface sink	-0.01	0 – -1
Total anthropogenic sources	*6.9*	*2.7 – 11.1*
Natural sources		
Soils under natural vegetation	6.6	3.3 – 9.0
Oceans	3.8	1.8 – 9.4
Lightning	–	–
Atmospheric chemistry	0.6	0.3 – 1.2
Total natural sources	*11.0*	*5.4 – 19.6*
Total natural + anthropogenic sources	**17.9**	**17.9 – 30.7**
Stratospheric sink	**14.3**	**4.3 – 27.2**

Denitrification

Denitrification (nitrate dissimilation) is the respiratory bacterial reduction of nitrate or nitrite to gaseous NO, N_2O, or N_2 (*Bremner*, 1997). It is generally accepted that denitrification is the main source for N_2O in agricultural soil *(Flessa* et al., 1998; *Dobbie & Smith*, 2001; *Khalil & Baggs*, 2005).
The pathway of denitrification is usually represented as follows:

Complete process: $N^{(+5)}\,O_3^- \rightarrow N^{(+3)}\,O_2^- \rightarrow N^{(+2)}\,O \rightarrow N_2^0$

Single steps:

(1) Nitrate reductase $NO_3^- + 2\ H^+ + 2e^- \rightarrow NO_2^- + H_2O$
(2) Nitrite reductase $NO_2^- + 2\ H^+ + e^- \rightarrow NO + H_2O$
(3) Nitric oxide reductase $2\ NO^- + 2\ H^+ + 2e^- \rightarrow N_2O + H_2O$
(4) Nitrous oxide reductase $N_2O + 2\ H^+ + 2e^- \rightarrow N_2 + H_2O$

During denitrification, N_2O is an obligate product and it is also possible to consume N_2O (*Firestone & Davidson*, 1989). Denitrification is mainly anaerobic, basically performed by aerobic bacteria that have the capacity to reduce N oxides (NO_3, NO_2, NO, N_2O) when O_2 becomes limiting (*Bremner*, 1997). The capacity to denitrify is widely spread among bacteria. *Pseudomonas* and *Alcaligenes* are the dominate species (*Tiedje*, 1988), but also some fungi and archaea can able to denitrify (*Hayatsu* et al., 2001). The main limiting factor for denitrification in mineral soils is the availability of organic material as electron donors for the reduction of nitrate. The proportion of N_2O/N_2 depends on the availability of reductants versus oxidants. N_2O will be produced if the availability of reductants is not sufficient. Among others, it strongly depends on soil pH, nitrate concentration and O_2 availability (*Firestone & Davidson*, 1989). Another pathway for N_2O production is the so-called co-denitrification. Here, denitrifying organism can use one N atom from nitric oxide (NO) or N_2O and combine it with an atom from another source, thus forming a hybrid product (*Baggs*, 2011).

Nitrification

Autotrophic nitrification (ammonia oxidation and nitrifier denitrification) is the oxidation of ammonium (NH_4^+) to nitrate (NO_3^-) by chemoautotrophic nitrifying bacteria (mainly by Nitrosomonas (reaction 1) and Nitrobacter (reaction 2)). This aerobic process is divided into two steps,

(1) $NH_4^+ + ⅔\ O_2 \rightarrow NO_2^- + H_2O + 2\ H^+$
(2) $NO_2^- + ½\ O_2 \rightarrow NO_3^-$

whereby NO and N_2O are intermediates (Figure 1.1). The NH_4^+ oxidizing bacteria only require CO_2, O_2 and NH_4^+ to proliferate. Beside the aforementioned aspects, limiting factors in soil include low pH, low water potential, phosphate availability, NO_2^- toxicity, and allelopathic compounds (*Bremner*, 1997). When oxygen supply is limited or soil moisture conditions are sub-optimal during nitrification, N_2O is produced due to the process called nitrifier denitrification by autotrophic nitrifying bacteria (*Wrage* et al., 2004, *Kool* et al., 2011). Another process, especially in acidic soils, is the heterotrophic nitrification of organic N (*Stange* et al., 2013) by heterotrophic bacteria and fungi. Because low pH the activity of autotrophic nitrifiers can be inhibit (*Weber & Gainey*, 1962).

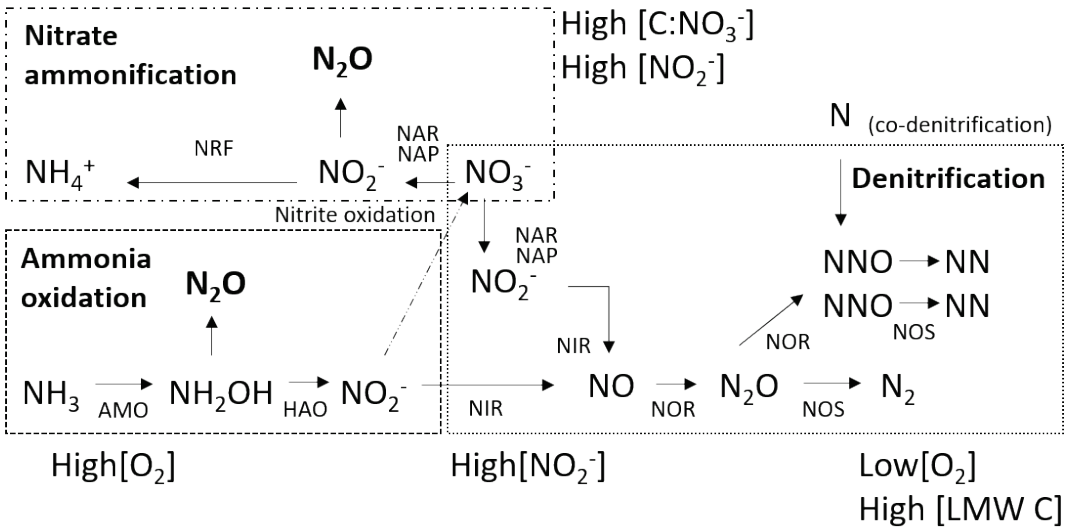

Fig. 1.1: Soil microbial pathways to N_2O production. LMW, low molecular weight; NO_3^-, nitrate; NO_2^-nitrite; NO, nitric oxide; NH_3, ammonia; NH_4^+, ammonium (adapted from *Baggs*, 2011).

Firestone & Davison (1989) described the result of all N_2O production and consumption by microbial processes in a simplified "hole-in-the-pipe" model (Figure 1.2). The two processes nitrification and denitrification are visualized as pipes with holes. N_2O production depends on the process rates, size of the leaks, as well as the diffusion and consumption of N_2O in the interaction of the soil and the atmosphere. The rate of N_2O production depends on enzymatic reactions, therefore many parameters like temperature, pH and water-filled pore space (WFPS) have a strong influence. The most important parameters are briefly discussed below.

Soil mineral N content

As substrates for nitrification and denitrification, soil ammonium and nitrate are inevitably associated with N_2O production. In countless studies, the relationship between N-input and direct N_2O emissions has been shown (*Bouwman*, 1996; *Flessa* et al., 1998; *Ruser* et al., 2001; *Bouwman* et al., 2002; *Walter* et al., 2015). N input in soil comes mainly from fertilization, atmospheric deposition of N-rich residues and mineralisation. In the past, it was believed that there is a linear relationship between N input and direct N_2O emissions, but recent studies show that increases in direct N_2O emission are related by a nonlinear relationship to increasing N input. Direct N_2O emissions increase abruptly at N input rates above plant uptake capacity (*Kim* et al., 2013). *IPCC* (2006) emission factors for fertilizers were calculated by the assumption of a linear relationship, although *Kim* et al. (2013) found a nonlinear relation. Further relationships between N and N_2O emissions are discussed in subsequent chapters.

Water-Filled Pore Space (WFPS)

N_2O production is strongly related to the WFPS, it includes information about the impact of soil water on aeration in addition to information on water availability (*Robertson & Groffman*, 2007). Depending on level, different microbial processes are predominant (Figure 1.3). Nevertheless, both nitrification and denitrification proceed simultaneously. It is well known that N_2O emissions rapidly increase, particularly after fertilization, if the WFPS is above 60 % (*Dobbie* et al., 1999; *Skiba & Ball*, 2002; *Batemann & Baggs*, 2005; *Ruser* et al., 2006). Above 60 % WFPS, O_2 availability is low and denitrification is the dominant process, but in soils with up to 60 % WFPS, nitrification can be a significant source for N_2O emissions (*Goreau* et al., 1980; *Linn & Doran*, 1984; *Abbasi & Adams*, 2000). The WFPS has also an effect on the final product of the reduction processes. In waterlogged soils, nitrate completely reduces to N_2. *Granli & Bøckmann* (1994), and *Davidson* (1992) found a relationship between WFPS and the formation of gaseous nitrogen (Figure 1.4). These threshold values can be variable with upward or downward shifts as shown in arable systems (*Bouwman* et al., 2002; *Ruser* et al., 2006). Extraordinarily high N_2O pulses in arable land can be caused by rewetting events after a dry period (*Ruser* et al., 2006).

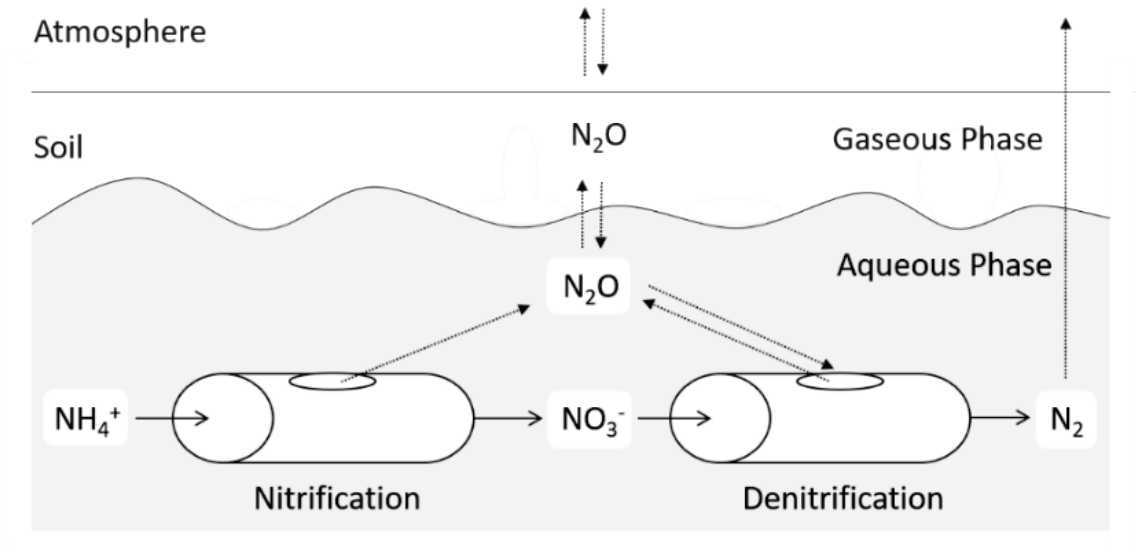

Fig. 1.2: Hole-in-the-pipe model, according to *Firestone & Davidson* (1989; modified).

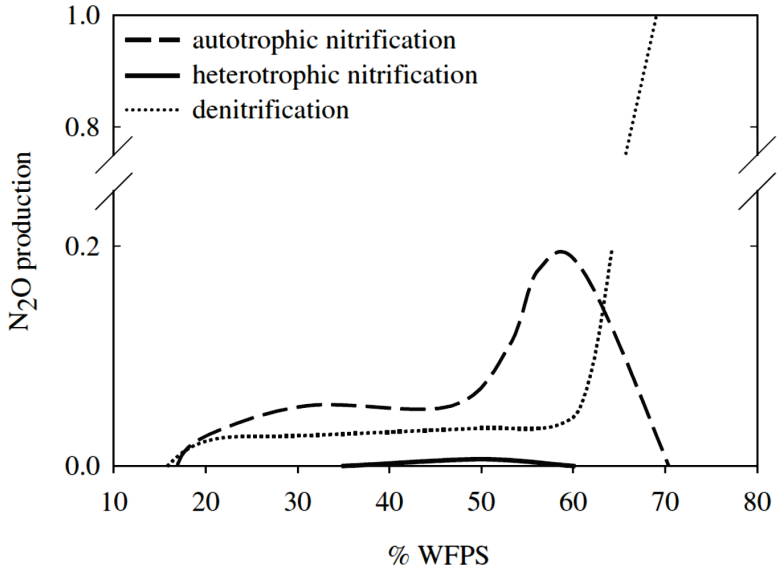

Fig. 1.3: N_2O production depending on WFPS of autotrophic and heterotrophic nitrification and denitrification (*Bateman & Baggs*, 2005).

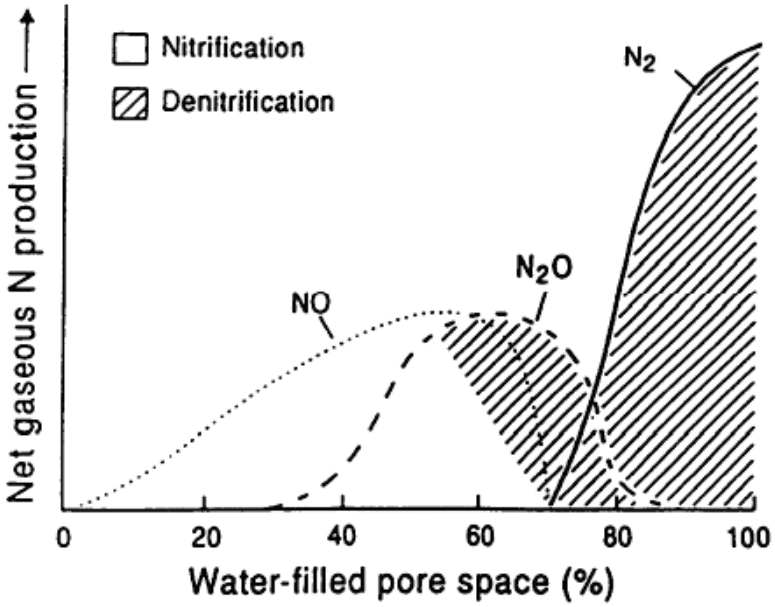

Fig. 1.4: Relationship between water-filled pore space and net production of nitric oxide (NO), nitrous oxide (N_2O) and dinitrogen (N_2) (*Davidson*, 1992; modified).

Tillage effect

Expected advantages of reduced tillage are the reduction of surface erosion by improving soil pore structure and stability (*Oades*, 1984) linked to increased water retention (*Lampurlanés* et al., 2001; *Copec* et al., 2015;) and C sequestration in the uppermost soil layer (*Alvarez*, 2005). Studies about tillage effects on N_2O emissions are contradictory. N_2O emissions are often higher in reduced or no till systems compared to conventional tillage with plough

practise as result of higher bulk density and water content, favouring conditions for denitrification (*Arah* et al., 1991; *Ball* et al., 1999; *Rochette* et al., 2008; *Hénault* et al., 2012; *Abdalla* et al., 2013). *Lognoul* et al. (2017) found 10 times larger emissions under a 7 year-old reduced tillage system compared to a conventional tillage system. This result was attributed to higher total N and soil organic carbon (SOC), and a larger bacterial biomass in the uppermost soil layer, caused by limited digging and mixing of crop residues under reduced tillage. However, there are other studies which measured lower N_2O emissions under reduced tillage (*Mutegi* et al., 2010; *Koga*, 2013; *Wang & Dalal*, 2015), or no differences between the treatments (*Chatskikh* et al., 2008; *Abdalla* et al., 2013; *Negassa* et al., 2015). In Europe reduced tillage is widespread due to its lower production costs and soil compaction (*Holland*, 2004). Further relationships between tillage practise and N_2O emissions are discussed in Chapter 7 and 9.

Crop residues

Global annual production of crop residues is approximately about 4 billion tons (*Lal*, 2005). Several studies reported high N_2O emissions from soil amended with crop residues (*Shan & Yan*, 2013; *Chen* et al., 2013; *Lehtinen* et al., 2014). Impacts of crop residues on N_2O emissions are shown in Figure 1.5. N_2O emissions caused by crop residues are strongly related to the C/N–ratio of the residues as well as the current WFPS, soil pH and texture (*Kaiser* et al., 1998; *Garcia-Ruiz & Baggs*, 2007; *Chen* et al., 2013). For estimates of N_2O emission, only the differences in N concentrations of residues are considered (*IPCC*, 2006). In a meta-analysis, *Chen* et al. (2013) found positive effects on soil N_2O emissions when C/N–ratios of crop residues were < 45, slightly positive effects for C/N–ratios of 45–100, and slightly negative effects for C/N–ratios >100. Turnover of crop residues by microorganisms can lead to oxygen depletion, which together with a high carbon availability can stimulate denitrification as one of the main processes for N_2O production. *Li* et al. (2016) found denitrification as main source of N_2O emission from residue-amended soils. Impact of OSR residues are discussed in detail in Chapter 7 and 8.

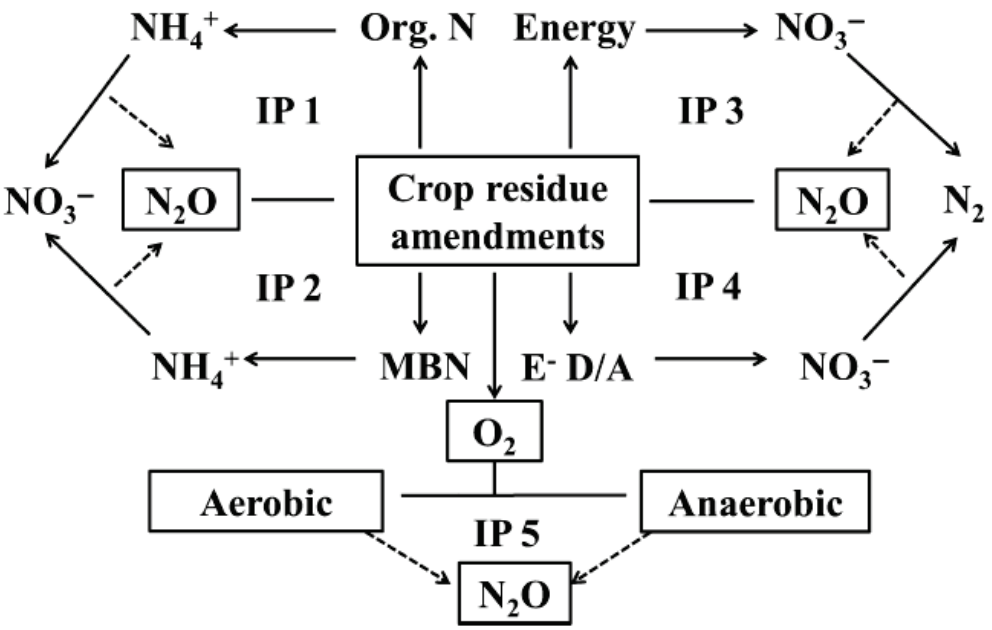

Fig. 1.5: Impacts of crop residue amendment on soil N_2O emissions. IP1 represents a positive effect due to crop residue input enhanced nitrification; IP2, a negative effect due to microbial N assimilation (MBN) - induced N limitation on nitrification; IP3, a positive effect due to energy supply for denitrification; P4, positive or negative effects depending on the relative abundance of electron donor and acceptor; and IP5, positive or negative effects depending on the level of soil anaerobicity (*Chen* et al., 2013).

2 Overview of Chapters and Aims

2.1 N_2O emissions from winter oilseed rape cultivation

Hypotheses: The emission factor of winter oilseed rape cultivation is overestimated. With a moderate N fertilization sufficient yield can be reached with N_2O emissions at a low level (Chapter 4)

Winter oilseed rape (*Brassica napus* L.) is the major oil crop cultivated in Europe. Rapeseed oil is predominantly used for production of biodiesel. The European Renewable Energy Directive requires that use of biofuels achieves GHG savings of at least 50 % compared to use of fossil fuel starting in 2018. N_2O field emissions are controlled by N fertilization and contribute up to 90 % to the GHG balance of winter oilseed rape cropping due to the high global warming potential of N_2O.

The main aims of this investigation were

(i) to determine direct annual N_2O emission from winter oilseed rape fields over a broad range of production sites in Germany, representing areas with a high proportion of winter oilseed rape within the crop rotations, thereby extending the currently available data for annual N_2O emissions from winter oilseed rape fields substantially,

(ii) to quantify the effect of N fertilization on N_2O fluxes and on yield–related N_2O emission, and

(iii) to deduce a fertilizer–related emission factor (FRE) specific for the production of winter oilseed rape

2.2 N_2O emissions affected by nitrification inhibitors

Hypothesis: Nitrification inhibitors mitigate nitrous oxide emissions in conjunction with organic (digestate) and mineral fertilization

Organic fertilization (Chapter 5)

Up to 90 % of the GHG emissions from biodiesel production can occur during OSR cultivation. Therefore, mitigation strategies for GHG emissions in the field are required and need to focus on direct nitrous oxide (N_2O) emission as one of the largest contributor. Substitution of synthetic N-fertilizers by digestates is currently under discussion due to the

avoidance of GHG emissions during the production process of synthetic fertilizer. Due to the sparse information on the effect of NIs on trace gas fluxes from OSR after digestate fertilization, the aim of this trial was to determine effects of NI application

(i) on N_2O emissions and mineral N dynamics
(ii) on grain and oil yield
(iii) on yield related emissions after digestate application in OSR production.

Mineral fertilization (Chapter 6)

As mentioned in the Chapter before, NI`s can have many advantages on the environment. To investigate the effect in treatments with mineral fertilizer, the following investigation was conducted.

The aim of the investigation was to determine the effect of the nitrification inhibitor DMPP (3,4–dimethylpyrazole phosphate) added to mineral fertilizer in oilseed rape production to evaluate

(i) the mitigation effect on direct N_2O emissions and
(ii) the effect of DMPP on oilseed rape grain yield

2.3 Contribution of OSR crop residues on N_2O emissions

Hypothesis: Winter oilseed rape residues contribute substantially on annual nitrous oxide emissions

Field study (Chapter 7)

Leaving crop residues in the field has many positive environmental effects such as nutrient transfer over winter, carbon sequestration, and reduction of soil erosion (*Chen* et al., 2013). On the other hand, adverse effects as, i.e., increased N_2O emissions after incorporation of crop residues were reported (*Baggs* et al., 2000; *Chen* et al., 2013). *Mosier* et al. (1998) estimated a global production of 0.4 million tons of N_2O-N y^{-1} from crop residues. Winteremissions can account for 50 % of the annual N_2O emissions if distinct frost/thaw cycles occur during this period (*Kaiser & Ruser*, 2000; *Jungkunst* et al., 2006). *Kaiser* et al. (1998) showed that N_2O emissions during the winter season decreased with increasing C/N–ratio of crop residues.
The effect of OSR crops residues on N_2O emissions during the post-harvest period have hardly been investigated, therefore the main hypotheses of this study were:

(i) as a result of incomplete immobilization of mineral N after harvest, N_2O emissions are stimulated through OSR crop residues which provide easily available C, which thus favour anaerobic conditions,

(ii) nevertheless, due to the high C/N–ratio of OSR residues, the emission factor derived from ^{15}N labelling technique of crop residues is lower than the IPCC default of 1 %, and
(iii) N_2O fluxes from reduced tillage system are higher than from the conventional system after OSR crop residue application

Incubation study (Chapter 8)

As complement to Chapter 7 an incubation study was conducted to gather more information about the effect of OSR residues on N_2O emissions.

The aim of the study was to investigate

(i) the effect of amount and quality of OSR residues on N_2O emissions, and
(ii) on NO_3^- losses

2.4 N_2O emissions effected by tillage

Hypothesis: Nitrous oxide emissions from a reduced tillage system are higher compared to conventional tillage (Chapter 9)

Reduced Tillage (RT) is distinguished by the facts of eliminating soil inversion and reducing soil disturbance by a shallow machining depth combined with conserving and managing crop residues (*Cunningham* et al., 2004). RT can increase the C sequestration in the uppermost soil layer (*Alvarez*, 2005) and reduce soil erosion by an increased water retention (*Lampurlanés* et al., 2001; *Copec* et al., 2015) as a result of improved soil pore structure and stability (*Oades*, 1984). The impact of RT on N_2O emissions can differ, studies reported higher (*Abdalla* et al., 2013; *Lognoul et* al., 2017), lower (*Koga*, 2013; *Wang & Dalal*, 2015) and similar N_2O emissions (*Abdalla* et al., 2010; *Negassa* et al., 2015).

This experiment investigated the effect of reduced tillage compared to conventional tillage on

(i) direct N_2O emissions, and the
(ii) effect on OSR grain yield

3 Materials and Methods

In this joint project, all basic treatments (No.1-7, and 10-12) were established at all sites (Table 3.1). Additional treatments to obtain more information about tillage (No.8) and crop residues effects (No.13-16) as well as effect of nitrification inhibitor with mineral fertilizer (No. 9) were additionally established in Hohenheim (Ihinger Hof; IHO). The experiment in Hohenheim was conducted at the research station Ihinger Hof, located 22 km west of Stuttgart, Germany (N 48° 44' 41''; E 8° 55' 26''). Figure 3.1 shows the location of all sites of the joint project on the map of Germany. The exact location of the study site at Ihinger Hof is shown in Figure 3.2. All sites and their main soil characteristics are shown in Table 3.3, bulk density before and after tillage is described in Table 3.2.

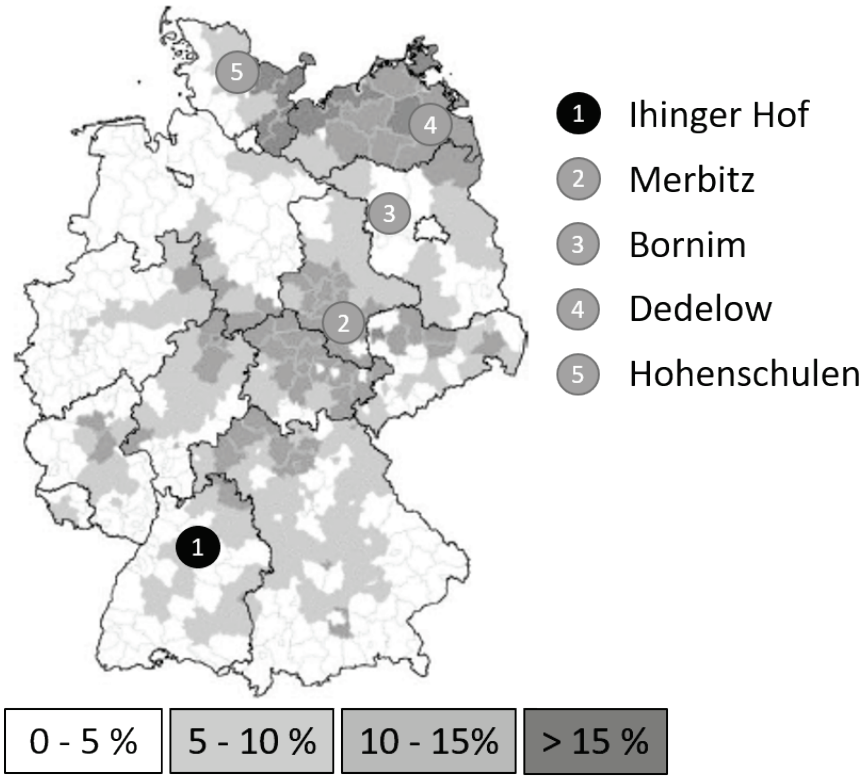

Fig. 3.1: Study sites of the joint project. Different grey intensity describes the percentage acreage of winter oilseed rape in German regions (*i.m.a.*, 2005, modified).

The greenhouse gas measurements were the main task of this study, including N_2O, CO_2 and CH_4 flux determination. Due to technical problems, the use of CH_4 data were unfortunately not possible. NH_3 measurement campaigns were conducted during the fertilization period, these data will be used of another working group of the project and not included in this thesis. Beside the GHG measurements, different soil data were collected (water content, bulk density, N_{min} content, and further chemical characteristics).

Plant quality like i.e. oil content of the seeds, plant height, biomass production, BBCH and C- and N- concentrations in crop residues was also monitored. Weather data from nearby climate station were provided by the experimental farm (air temperature in 0.5, 1 and 2 m height, wind speed and direction, and precipitation).

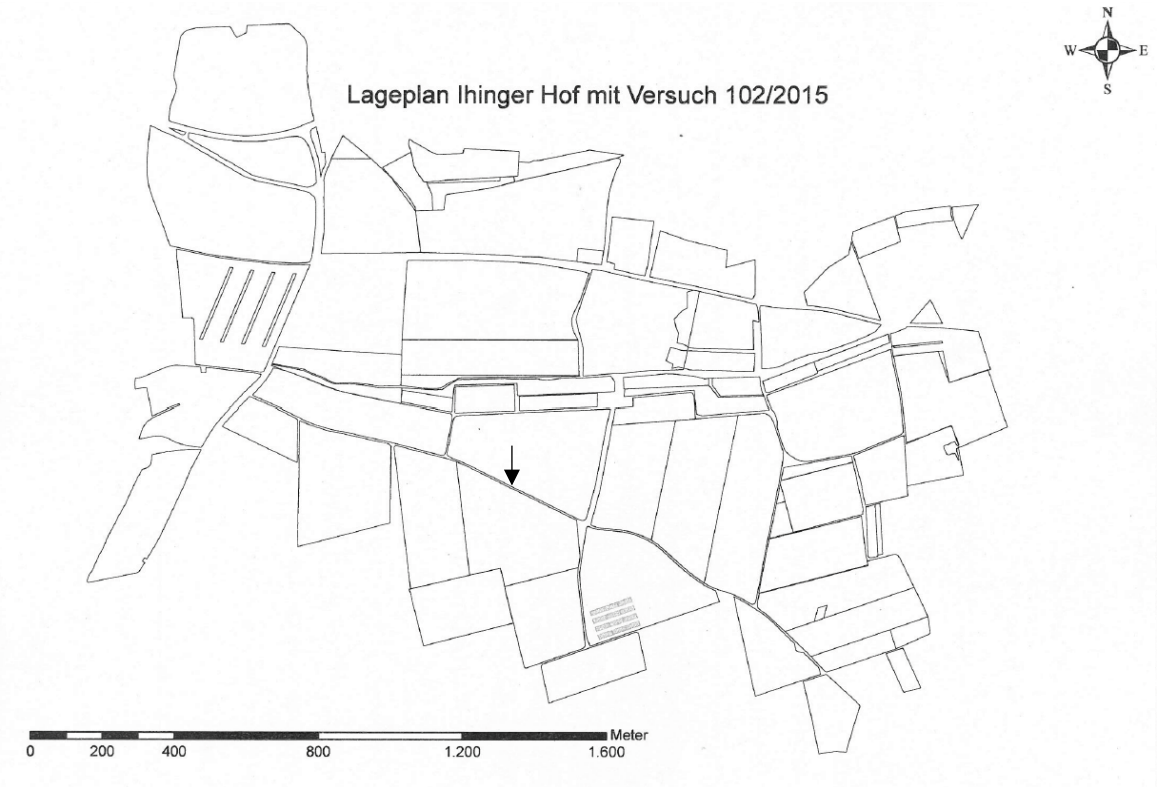

Fig. 3.2: Map of the research station Ihinger Hof; study site indicated by an arrow (provided by research station Ihinger Hof, 2017).

Tab. 3.1: Overview of the treatments in this study: site (all: all sites of the joint project; IHO: Ihinger Hof), GL: grassland, fertilizer (CAN: calcium ammonium nitrate; ASN: ammonium sulphate nitrate, level of N-fertilizer, nitrification inhibitor (NI; DMPP: 3,4-dimethylpyrazole phosphate; TZ+MP: 1H-1,2,4-triazole and 3-methylpyrazole) and catch crop type.

No	Site	Treatment	N-fertilizer	N-Amount	NI	Catch crop	Chapter with further information
1	all	N1	-	0	-	OSR	4
2	all	N2	CAN	60	no	OSR	4
3	all	N3	CAN	120	no	OSR	4
4	all	N4	CAN	180	no	OSR	4;6;9
5	all	N5	CAN	240	no	OSR	4
6	all	N6	Digestate	180	no	OSR	5
7	all	N7	Digestate	180	TZ+MP	OSR	5
8	IHO	RT	CAN	180	no	OSR	9
9	IHO	ENTEC	ASN	180	DMPP	OSR	6
10	all	N10	CAN	180	no	WB	4
11	all	N11	CAN	180	no	WW	4;5
12	all	GL	-	0	-	-	-
^{15}N-experiment							
13	IHO	CT +CR	CAN	180	no	WW	7
14	IHO	CT -CR	CAN	180	no	WW	7
15	IHO	RT +CR	CAN	180	no	WW	7
16	IHO	RT -CR	CAN	180	no	WW	7

At each site, a randomized split-plot experiment with four replicated blocks was established in 2012 (Figure 3.3). An example for one plot is shown in Figure 3.4. Crop rotation was identical over all sites. All crops of the rotation were cultivated as main plots in each of the four blocks described in detail in Chapter 4. These included: winter oilseed rape (var. 'Visby'), winter wheat (var. 'Julius'), and winter barley (var. 'Tenor' in Bornim, var. 'Meridian' in Hohenschulen and var. 'Souleyka' at all other sites).

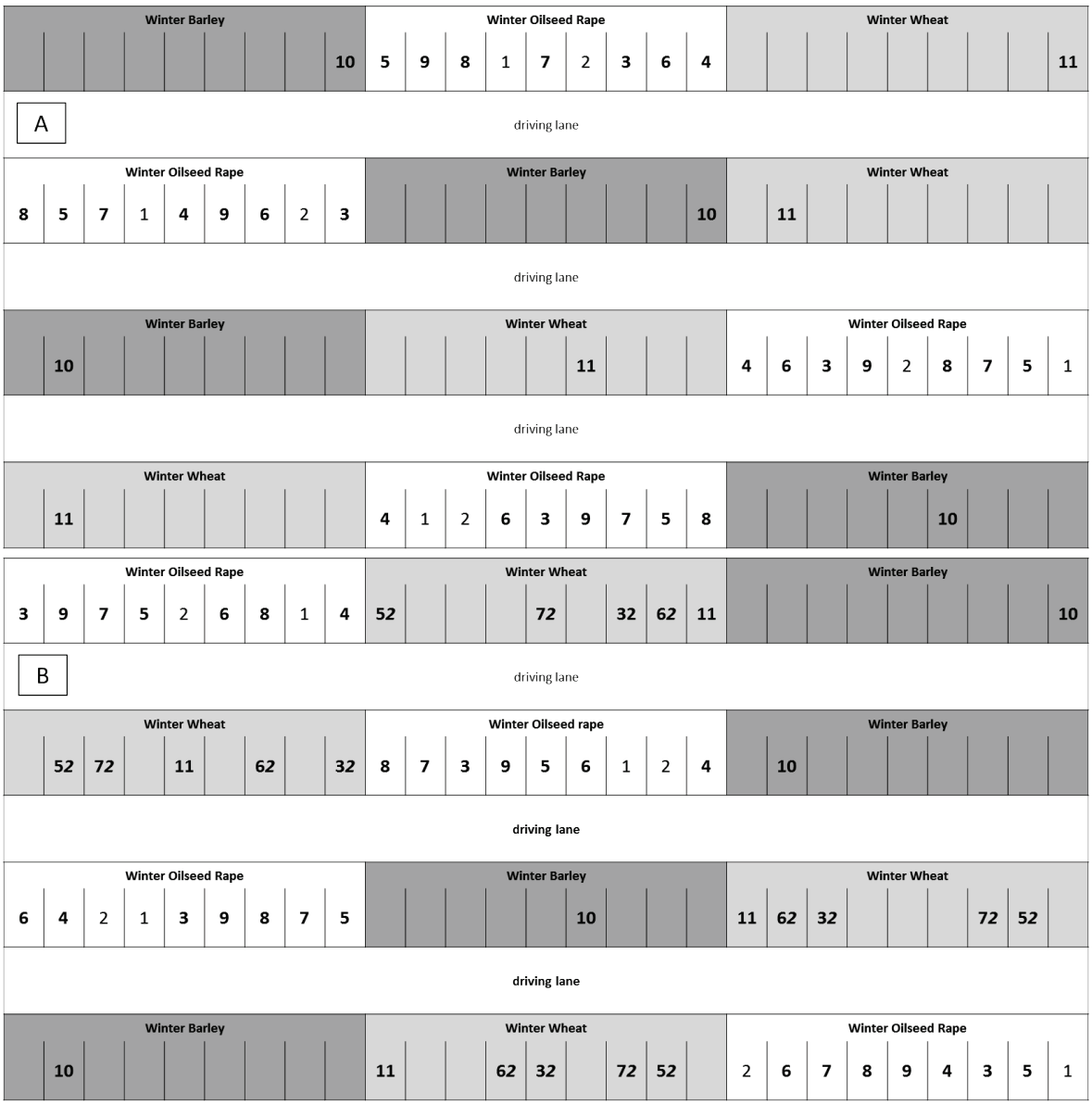

Fig. 3.3: Randomized split-plot experiment at Ihinger Hof. A: Field plan 2012 with sampled treatments, plots on which gas emissions were measured are labelled with bold numbers. B: Field plan 2013, additional treatments in winter wheat to investigate the post-harvest effect of winter oilseed rape.

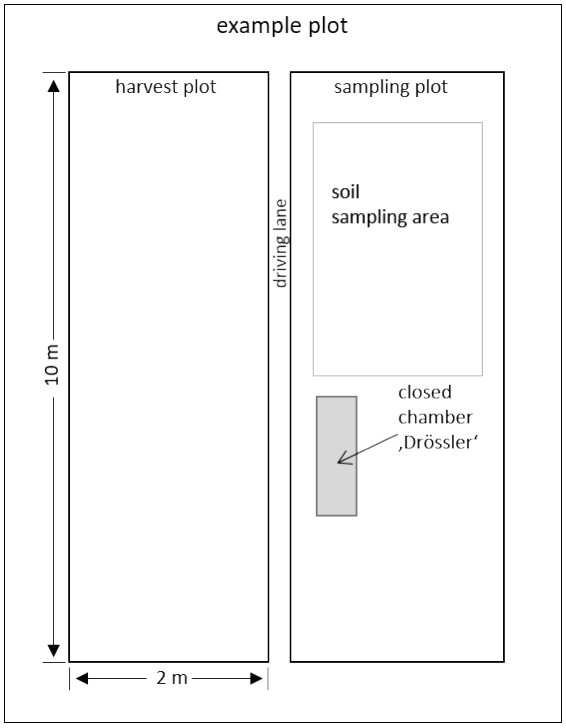

Fig. 3.4: Experimental plot constituting an undisturbed subplot for harvest to determine yield and a sampling subplot for gas measurement, soil sampling and biomass samples.

Tab. 3.2: Mean (n = 4, ± SE) soil bulk density (0 – 30 cm depth)as affected by tillage during the investigation period. Soil was sampled from the same treatments where gas samples were taken.

	01.08.13 before tillage		16.12.13 after tillage		31.07.14 before tillage		26.02.15 after tillage		18.08.15 before tillage	
					g m^{-3}					
Treatment	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
N3	1.20	0.04	1.39	0.03	1.40	0.01	1.20	0.03	1.21	0.06
N4	1.17	0.04	1.39	0.03	1.42	0.06	1.24	0.05	1.20	0.05
N5	-	-	-	-	1.34	0.05	1.27	0.07	1.11	0.05
N6	1.22	0.02	1.38	0.05	1.33	0.08	1.21	0.05	1.27	0.08
N7	1.20	0.03	1.33	0.09	1.38	0.05	1.26	0.03	1.21	0.10
RT	1.23	0.01	1.46	0.03	1.40	0.04	1.31	0.04	1.27	0.03
ENTEC	1.20	0.03	1.39	0.03	1.31	0.09	1.25	0.01	1.23	0.06
N10	1.25	0.06	1.25	0.06	1.25	0.07	1.28	0.03	1.23	0.01
N11	1.28	0.04	1.31	0.02	1.27	0.07	1.21	0.04	1.20	0.08

Tab. 3.3: Main soil characteristics of all study sites.

Study site		Bornim	Dedelow	Hohenheim	Hohenschulen	Merbitz
Coordinates		N 52° 37' 0" E 12° 46' 60"	N 53° 21' 57" E 13° 49' 38"	N 48° 44' 41" E 8° 55' 26"	N 54° 18' 48" E 9° 59' 36"	N 51° 36' 58" E 11° 91' 12"
MAP	[mm a^{-1}]	503	485	688	732	520
2013/2014/2015		615/482/570	446/561/414	923/763/544	462/409/562	700/456/429
MAT	[°C]	8.7	8.4	8.3	8.9	9.0
2013/2014/2015		9.4/13.0/10.6	8.7/9.9/9.7	8.6/10.4/10.1	8.1/9.6/8.8	9.1/10.7/10.4
Soil type (IUSS, 2015[$])		Luvisol	Luvisol	Haplic Luvisol	Haplic Luvisol/Anthrosol	Haplic Chernosem
Clay	[%]	5.7	10.0	3.2	10.5	15.8
Silt	[%]	19.9	30.9	78.2	29.4	67.8
Sand	[%]	74.4	59.1	18.6	60.1	16.4
pH[§]	0.01 *M*	6.5	7.4	6.8	5.9	6.6
C_{org}[§]	[%]	1.15	0.75	1.68	1.87	1.18
N_t[§]	[%]	0.09	0.10	0.20	0.12	0.11

MAP: Long-term mean annual precipitation and annual precipitation in the single experimental years; MAT: Long-term mean annual air temperature (2m) and annual mean air temperature in the single experimental years, [§] measured in the top soil (0-30 cm)
[$] IUSS Working Group WRB (2015)

4 N_2O emissions from winter oilseed rape cultivation

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Nitrous oxide emissions from winter oilseed rape cultivation

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ABSTRACT

Winter oilseed rape (*Brassica napus* L., WOSR) is the major oil crop cultivated in Europe. Rapeseed oil is predominantly used for production of biodiesel. The framework of the European Renewable Energy Directive requires that use of biofuels achieves GHG savings of at least 50% compared to use of fossil fuel starting in 2018. However, N_2O field emissions are estimated using emission factors that are not specific for the crop and associated with strong uncertainty. N_2O field emissions are controlled by N fertilization and dominate the GHG balance of WOSR cropping due to the high global warming potential of N_2O. Thus, field experiments were conducted to increase the data basis and subsequently derive a new WOSR-specific emission factor.

N_2O emissions and crop yields were monitored for three years over a range of N fertilization intensities at five study sites representative of German WOSR production. N_2O fluxes exhibited the typical high spatial and temporal variability in dependence on soil texture, weather and nitrogen availability. The annual N_2O emissions ranged between 0.24 kg and 5.48 kg N_2O-N ha^{-1} a^{-1}. N fertilization increased N_2O emissions, particularly with the highest N treatment (240 kg N ha^{-1}). Oil yield increased up to a fertilizer amount of 120 kg N ha^{-1}, higher N-doses increased grain yield but decreased oil concentrations in the seeds. Consequently oil yield remained constant at higher N fertilization. Since, yield-related emission also increased exponentially with N surpluses, there is potential for reduction of the N fertilizer rate, which offers perspectives for the mitigation of GHG emissions.

Our measurements double the published data basis of annual N_2O flux measurements in WOSR. Based on this extended dataset we modeled the relationship between N_2O emissions and fertilizer N input using an exponential model. The corresponding new N_2O emission factor was 0.6% of applied fertilizer N for a common N fertilizer amount under best management practice in WOSR production (200 kg N ha^{-1} a^{-1}). This factor is substantially lower than the linear IPCC Tier 1 factor (EF1) of 1.0% and other models that have been proposed.

1. Introduction

In the context of biofuel production especially nitrous oxide (N_2O) contributes to high GHG emissions during the step of biomass production (Dufossé et al., 2013; Hong, 2012). N_2O is a climate relevant trace gas that absorbs light in the IR spectrum and therefore reduces the atmospheric transparency to thermal radiation from the earth's surface (Granli and Bøckman, 1994). The atmospheric N_2O concentration in the last decade increased by 0.73 ppb a^{-1} and with a mean concentration of 328 ppb in 2015 it exceeded the pre-industrial level by about 21%

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(WMO, 2016). N_2O contributes 7.4% (0.17 W m^{-2}) of the total anthropogenic radiative forcing (IPCC, 2013); it has a high heat adsorption capacity, a long atmospheric lifetime of more than 100 years and has a 296 fold higher global warming potential (IPCC, 2001; RED, 2009) compared to the same mass of carbon dioxide (CO_2). Besides its contribution to the greenhouse effect, N_2O also contributes to stratospheric ozone depletion (Crutzen, 1981; Ravishankara et al., 2009).

Approximately 60% of anthropogenic N_2O emissions are released by agricultural soils (Clais et al., 2013). There is general agreement that nitrification and biological denitrification are the main sources for N_2O production in soils (Bremner, 1997), whereas the contribution of other processes such as nitrifier-denitrification is currently under discussion (Wrage et al., 2001; Shaw et al., 2006; Butterbach-Bahl et al., 2013).

All processes of N_2O production in soils rely on mineral N (i.e. Ruser et al., 2001; Zebarth et al., 2008). Therefore, N_2O emissions from agricultural soils generally increase with increasing N fertilization as it provides the substrates (NO_3^-, NH_4^+) for N_2O production (i.e. Stehfest and Bouwman, 2006). Furthermore, N_2O emission is correlated with N surpluses (N fertilization – N removal) in arable systems (Kaiser and Ruser, 2001; Van Groenigen et al., 2004) as well as in horticultural systems (Pfab et al., 2011).

Oilseed rape (*Brassica napus* L.) is the major oil crop in Europe, accounting for more than 70% of the European oilseed volume in 2012 (Carré and Pouzet, 2014). In 2014, oilseed rape covered 9.1 × 10^6 ha or approximately 8.5% of the total European arable land (FAO, 2016). The corresponding mean grain yield was 3.17 Mg ha^{-1}. In the same year, the mean grain yield in Germany was 4.48 Mg ha^{-1} on 1.4 × 10^6 ha (German Federal Statistical Office, 2017), showing both the high potential for winter oilseed rape (WOSR) cultivation as well as the reason for Germany's leading position (together with France) regarding WOSR production in the EU.

The acreage of WOSR in the European Union more than doubled between 2003 and 2014 (FAO, 2016), which went along with the increase of biodiesel contributing more than 75% of the transport biofuels in Europe (Hamelinck et al., 2013). This increased production is also a result of the Renewable Energy Directive (RED, 2009), in which the European Union mandates a share of 10% from renewables in the transport energy sector by 2020. The RED also defined sustainability criteria for biofuels, which were updated in 2015 (EU, 2015). According to these criteria, biofuels can only be considered and consequently subsidized as such if they contribute to a total reduction of greenhouse gas emissions (GHG) of 35% (current reduction value) and, starting from 2018, of 50% (for production plants that became operational before October 2015) and by 60% (for new production plants) in comparison to the use of fossil fuel.

WOSR is a crop demanding high amounts of N fertilizer to build up efficient photosynthetic leaf tissue (Hegewald et al., 2016). Maximum yields are often achieved with N rates exceeding 200 kg N ha^{-1} whereas N removal with the seeds as well as the N harvest index are low, thereby resulting in high N surpluses of up to 90 kg N ha^{-1} a^{-1} (Henke et al., 2007; Sieling and Kage, 2010). It has also been reported that large amounts of crop residues (petals and leaves), which can be mineralized easily, are returned to the soil after flowering (Sieling and Kage, 2010). Furthermore, N uptake by WOSR plants ends early and increases in N content in seeds during pod filling is more the result of N translocation from vegetative plant parts than from N uptake from soil (Malagoli et al., 2005); both will result in enhanced soil mineral N contents during or shortly after the harvest period. Winter wheat (*Triticum aestivum* L.) is the predominant succeeding crop for WOSR in German crop rotations. The N uptake of winter wheat before winter is approximately 20 kg N ha^{-1} and as such markedly below the N release after WOSR cultivation (Sieling and Kage, 2010). Both, the N surpluses as well as the high soil nitrate contents have the potential of fueling N_2O production in soils.

Due to the high global warming potential of N_2O, the assessment of N_2O emissions with a reliable emission factor is of vital importance for the calculation of GHG balances of biofuels, such as biodiesel produced from WOSR. Results from life cycle analysis (LCA) suggest that direct and indirect N_2O emissions account for between 20 and 40% of the total GHG emission associated with the production and consumption of biodiesel (Hong, 2012; Dufossé et al., 2013). For a bioethanol production system, the choice of different available N_2O emission factors in LCAs might result in completely contrasting results and conclusions, as Smith and Searchinger (2012) remarkably demonstrated. Following IPCC guidance, they set the emission factor to 1.5% (including direct and indirect emissions) and the corresponding emission reached the 35% GHG reduction goal. Using the distinct higher emission factor of 4%, as suggested by Crutzen et al. (2008), based on their so called "top-down" approach, the reduction potential for wheat-based bioethanol was completely eliminated.

In order to assess fertilizer-induced N_2O emissions, different N_2O emission factors have been proposed. The IPCC (2006) guidelines suggest a constant direct N_2O-N loss of 1% of N applied and N in crop residues. This default emission factor was modified from a global data set for wheat and grassland sites originally provided by Bouwman (1996) and, as mentioned by Bouwman, does not consider crop type or site-specific effects. A further drawback of this emission factor is that N_2O emissions do not necessarily correlate linearly with N fertilizer amounts and that N_2O emissions increase over-proportionally when high N fertilizer doses exceed plant demand (McSwiney and Robertson, 2005; Hoben et al., 2011; Kim et al., 2013).

The Joint Research Centre (JRC) of the EU provides an online tool (the so-called Global Nitrous Oxide Calculator, GNOC) to assess GHG emissions from biofuels in EU legislation (Edwards et al., 2013). This tool calculates N_2O emissions based on the approach of Stehfest and Bouwman (2006). It uses an exponential algorithm that considers site and management specific characteristics such as soil texture, climate, soil organic matter, pH and vegetation. In this model, WOSR was originally in the vegetation class "other" but the JRC recently moved it into the same class as "cereals" without refitting the model (Edwards et al., 2016). This resulted in a calculative reduction of the N_2O emissions from WOSR.

The decision to move WOSR to the cereals group in the GNOC tool is supported by Walter et al. (2015) who used data sets on N_2O emissions from WOSR fields to run a meta-analysis. They also used an exponential model for fertilizer-derived N_2O emission from WOSR, which resulted in even lower N_2O emissions than the GNOC tool.

In regions with strong frost-thaw cycles, high N_2O fluxes can occur during thawing periods (Flessa et al., 1995; Röver et al., 1998). These high thaw pulses can account for more than 50% of the annual N_2O budget from agricultural soils (Kaiser and Ruser, 2001; Jungkunst et al., 2006). Due to these high N_2O winter fluxes, annual measurements are a prerequisite for the reliable quantification of N_2O emissions. Consequently, the duration of the period of trace gas measurements was a criterion for the inclusion (measurements covering > 300 days) or exclusion of data sets in the review by Walter et al. (2015), and only 12 studies with 18 annual datasets (43 data points in total) fulfilled this criterion. Additionally, the small dataset showed a high variability of the N_2O emissions among study sites and also among experimental years.

The main aims of our investigations were therefore: (i) to determine direct annual N_2O emission from WOSR fields over a broad range of production sites, representing areas with a high proportion of WOSR within the crop rotations, thereby extending the currently available data substantially, (ii) to quantify the effect of N fertilization on N_2O fluxes and on yield-related N_2O emission, and (iii) to deduce a fertilizer-related emission factor (FRE) specific for the production of winter WOSR.

2. Materials and methods

2.1. Study sites, experimental design and management

Trace gas measurements were conducted at five study sites located

Table 1
Meteorological, soil chemical and physical characteristics of the study sites.

Study site	Coordinates	MAP 2013/2014/2015	MAT 2013/2014/2015	Soil type (IUSS, 2015[$])	Soil texture[§]			pH[§]	C_{org}[§]	N_t[§]
		[mm a^{-1}]	[°C]		Clay [%]	Silt [%]	Sand [%]	0.01 *M* $CaCl_2$	[%]	[%]
Berge	N 52°37′0″	503	8.7	Luvisol	5.7	19.9	74.4	6.5	1.15	0.09
	E 12°46′60″	615/482/570	9.4/13.0/10.6							
Dedelow	N 53°21′57″	485	8.4	Luvisol	10.0	30.9	59.1	7.4	0.75	0.10
	E 13°49′38″	446/561/414	8.7/9.9/9.7							
Ihinger Hof	N 48°44′41″	688	8.3	Haplic Luvisol	3.2	78.2	18.6	6.8	1.68	0.20
	E 8°55′26″	923/763/544	8.6/10.4/10.1							
Hohenschulen	N 54°18′48″	732	8.9	Haplic Luvisol/Anthrosol	10.5	29.4	60.1	5.9	1.87	0.12
	E 9°59′36″	462/409/562	8.1/9.6/8.8							
Merbitz	N 51°36′58″	520	9.0	Haplic Chernosem	15.8	67.8	16.4	6.6	1.18	0.11
	E 11°91′12″	700/456/429	9.1/10.7/10.4							

MAP: Long-term mean annual precipitation and annual precipitation in the single experimental years; MAT: Long-term mean annual air temperature (2 m) and annual mean air temperature in the single experimental years.
[§] measured in the top soil (0–30 cm).
[$] IUSS Working Group WRB (2015).

in representative areas with a high share of WOSR in the crop rotation. Three sites were located in (1) northern Germany representing approximately 55% of the total German WOSR production area (Hohenschulen, University Kiel; Dedelow, ZALF Müncheberg; Berge, ATB Potsdam-Bornim/Humboldt University Berlin), (2) one site in central Germany (Merbitz, University Halle-Wittenberg) representing 30%, and (3) one site in southern Germany (Ihinger Hof, University Hohenheim) representing 15% of the total German oilseed area. The main characteristics of the study sites are shown in Table 1.

At each site, a randomized split-plot experiment with four replicated blocks was established in 2012. The crop rotation was identical at all sites. All crops of the rotation, winter oilseed rape (var. 'Visby'), winter wheat (var. 'Julius'), and winter barley (*Hordeum vulgare* L., var. 'Tenor' in Berge and var. 'Souleyka' at all other sites), were cultivated as main plots in each of the four blocks. Within the WOSR main plots, at least seven different treatments were included, whereas the main plots with winter wheat and winter barley were managed according to best agricultural management practices without any further differentiation within the crop. Plot size varied slightly over the study sites due to different farming machinery; the minimum size was 3 × 9 m (27 m^2).

WOSR was sown at all sites between end of August and the first two weeks in September (40–45 grains m^{-2}, inter-row width was 0.36 m). In early spring, 90 kg S ha^{-1} were applied as kieserite ($MgSO_4 \cdot H_2O$) to avoid S deficiency in all WOSR treatments including the unfertilized control. After harvest, in the period between mid-July and early August, the soil was ploughed to a depth of 25 cm and winter wheat was subsequently sown at the end of September or in early October. Crop protection and further management measures were conducted according to site-specific agricultural practice. At Berge WOSR straw was removed after harvest whereas it remained on the field at the other study sites. This removed about 20 kg N ha^{-1} a^{-1} (2.8 Mg C ha^{-1} a^{-1}, C/N = 105) from the site Berge (median of all years and treatments).

The treatments of WOSR relevant for results described hereafter, were an unfertilized control and treatments fertilized with 60, 120, 180, or 240 kg N ha^{-1} a^{-1} for yield determination. Typical WOSR fertilization targets in Germany are in the range from 180 kg ha^{-1} a^{-1} to 210 kg ha^{-1} a^{-1}. The 180 kg N ha^{-1} a^{-1} fertilization treatment represents a typical fertilization target value of 200 kg N subtracting N_{min} contents after winter (approximately 20 kg N ha^{-1}). At all sites we measured N_2O fluxes also in additional treatments such as biogas residue application or soil tillage variants. However, these will be discussed in subsequent publications. Trace gas fluxes were measured in every year and at every site in the 120 kg N ha^{-1} and in the 180 kg N ha^{-1} treatment and in some further N treatments (including some of the unfertilized controls) in single years (Table 2). N fertilization to WOSR was split into two equal doses with a first application at the beginning of the growing season and the second application in BBCH-stage 5 (inflorescence emergence, Meier, 2001) approximately four weeks after the first N application. We used calcium ammonium nitrate (CAN) for all N applications.

2.2. Flux measurements

Using the closed chamber method (Hutchinson and Mosier, 1981), flux measurements were conducted at least once a week starting in January or February 2013 and ending in December 2015. Chambers were placed between the seed rows of WOSR, but included the plants for measurements in cereals. In order to place the chambers between the plant rows, the chambers' dimensions were 71 cm length, 27 cm width and 10 cm height. Chamber material was white opaque PVC (PS-plastic, Eching, Germany). They were equipped with rubber sealing, a pressure vent and a ventilator. For measurements, the chambers were anchored on their frames using elastic straps. The frame height was

Table 2
Median of all measured N_2O flux rates as affected by study site, experimental year (Exp. Year) and N fertilization.

Study site	Exp. year	N fertilization [kg N ha^{-1} a^{-1}]			
		0	120	180	240
		N_2O flux [µg N_2O-N m^{-2} h^{-1}]			
Berge	2013	–	2.2	1.3	–
	2014	–	0.7	0.9	–
	2015	–	1.9	1.1	1.3
	2013–2015[$]	–	1.6	1.1	–
Dedelow	2013	–	2.4	2.9	–
	2014	2.2	3.2	3.2	–
	2015	1.9	2.9	2.6	3.0
	2013–2015[$]	–	2.8	2.9	–
Ihinger Hof	2013	–	4.3	8.8	–
	2014	–	5.3	5.3	8.9
	2015	–	1.6	1.7	3.6
	2013–2015[$]	–	3.7	5.3	–
Hohenschulen	2013	2.5	4.8	7.3	–
	2014	3.0	5.2	6.3	–
	2015	–	9.0	8.1	14.2
	2013–2015[$]	–	6.3	7.3	–
Merbitz	2013	6.3	8.7	13.0	–
	2014	4.7	14.2	12.5	–
	2015	3.7	5.9	5.7	7.8
	2013–2015[$]	4.9	9.6	10.4	–

-not determined/not calculated.
[$] Mean values (only given for treatments with 3 years of measurements).

13 cm and they were installed in soil to a depth of 5 to 10 cm. During flux measurements, the chambers were kept closed for one hour; gas samples were taken every 20 min using vacutainers or stopcock vials, resulting in four gas samples per flux measurement. Chamber temperature was recorded for each gas sample. Gas samples were analyzed for N_2O and CO_2 concentrations in the laboratories of the participating research groups by various gas chromatographs equipped with electron capture and flame ionization detectors as well as automatic samplers. Lab inter-comparability was verified by conducting blind inter-comparison measurements between the labs in the beginning of the study. Each laboratory achieved a coefficient of variance below 2% on ten repeated measurements of an ambient N_2O standard gas (data not shown).

2.3. Environmental, soil, and plant analyzes

A climate station was installed next to the experimental plots at each of the study sites. We measured precipitation and air temperature (2 m and 5 cm height). Additionally, soil temperature in one of the four replicated main plots was recorded in 5, 10, and 20 cm soil depth (Logtacs, TRIX-8, CIK solutions, Karlsruhe, Germany).

Simultaneously to each gas sampling, soil samples were taken from 0 to 30 cm depth with an auger. The soil from three insertions per replicate plot was pooled over the four replicates, sieved ($<$ 5 mm) and stored frozen until further analysis. Additionally, in early spring and after harvest of the WOSR N_{min} was determined in 0–30, 30–60, and 60–90 cm depth. These soil samplings were carried out for each plot separately.

For the quantification of mineral N contents, 80 g of soil were extracted with 200 ml of a 0.0125 *M* $CaCl_2$ solution. Concentrations of NO_3^- and NH_4^+ in the extracts were determined using flow-injection analyzers. The analyzers used for that purpose were tested for comparability in an inter-laboratory test. A further aliquot of the soil was used to determine soil moisture by drying at 105 °C for one day.

Before and after soil management events, bulk density of the top soil was determined using stainless steel cylinders (100 ml).

Fresh matter yield was determined by cutting WOSR plants from 1 m^2. The green cut was separated into straw and pods which were flailed subsequently. Moisture was determined after drying for three days at 60 °C. Aliquots of the milled straw and grains were analyzed for C and N using an elemental analyzer (vario Max CN, Elementar Analysensysteme, Hanau, Germany). The oil content of the rapeseeds was determined with NIRS (NIRSystem 5000, Foss, Hamburg, Germany).

2.4. Calculations and statistical analyzes

2.4.1. Flux calculation

Molar gas concentrations were transformed into mass concentrations according to the ideal gas law taking chamber temperature and standard pressure into account. We used several criteria to select the most appropriate flux calculation model and to evaluate the reliability of calculated fluxes. The Akaike information criterion (AIC) was used to decide between flux calculation by the HMR model (Pedersen et al., 2010) and robust linear regression (Huber, 1981). The HMR estimate was used if its AIC value was smaller than the AIC from linear regression and if its kappa value, which controls the curvature, was smaller than 20 h^{-1}. Restricting kappa this way avoids strong overestimation of fluxes due to outliers of the first concentration – time point, which can result in an excellent fit of the nonlinear model but extreme curvatures and flux estimates. A linear regression was applied if only three gas samples were available for flux determination.

The resulting gradients at time zero were multiplied with chamber volume divided by chamber area to derive the flux estimates. For this, the height of the frame was determined after any changes, such as reinstallation after tillage measures. Snow was considered part of the soil and not part of the chamber headspace.

We used the generally clear and significant increase of CO_2 concentration in closed chamber at temperatures above the freezing point to check for accurate diffusive gas accumulation, which can be affected in particular by high wind speed and changing pressure conditions (Hutchinson and Mosier, 1981; Hutchinson and Livingston, 2001). Measured fluxes were subjected to a rigorous quality check since many different people were involved in the comprehensive gas sampling (about 60,000 gas samples were taken in the whole project during the three experimental years) and occasionally different anchoring of the chambers could have resulted in small leakages at the rubber sealing. Thus, the Pearson correlation coefficient between CO_2 concentration and closing time was used as an indicator of the reliability of diffusive gas accumulation. If the Pearson coefficient of the CO_2 flux was smaller than +0.85 and air temperature was above 0 °C, we rated the calculated diffusive flux as considerably biased by other processes and the corresponding flux measurement was excluded from analyses.

Only a few measurements indicated exceptionally high N_2O uptake rates of more than 100 μg N m^{-2} h^{-1}. These fluxes, which were usually associated with abnormally high initial N_2O concentrations, were considered unreliable given that negative diffusion gradients are limited due to low concentration in the atmosphere. Single N_2O flux estimates with extraordinary high uncertainty, i.e., standard errors above 100 μg N m^{-2} h^{-1} (above 25 μg N m^{-2} h^{-1} for uptakes larger than 50 μg N m^{-2} h^{-1}) were also rated as highly unsure and removed. The 90% quantile of all flux standard errors was 11 μg N m^{-2} h^{-1} (median: 1.5 μg N m^{-2} h^{-1}). Thus, the flux detection limit was lower than approximately 2 * SE = 22 μg N m^{-2} h^{-1} for 90% of the flux measurements.

After these rigorous quality checks approximately 10,000 measured N_2O flux rates were analyzed further for the results presented hereafter. Only a very small number of sampling dates were completely lost through the quality check but it resulted in some sampling dates with a reduced number of replicates. Multiple imputation (Honaker et al., 2011) was employed to fill these gaps in plot specific N_2O time series for subsequent statistical analysis. Imputation was done between the replicate N_2O time series group-wise by sites, treatments and years. To improve the performance of imputation, data of each group was transformed as log(flux − min(flux) + 1). The number of multiple imputations was set to 25 and linear time effects were used to account for autocorrelation. The imputed fluxes were then transformed back and cumulated fluxes calculated by linear interpolation. Finally, the median of the multiple imputations was calculated and used as cumulated flux estimate.

2.4.2. Cumulative N_2O emissions and emission factor

For the calculation of annual N_2O emissions we cumulated N_2O fluxes between 1st January and 31st December for each experimental year. We defined this time period at the beginning of our investigations based on the following expectations: except for the sowing, this period covered all soil management and N fertilization measures of WOSR cultivation as well as the time period with presumably increased soil mineral N contents following WOSR harvest and under subsequent winter wheat. Since there was no N fertilization in autumn, we did not expect increased fluxes after rapeseed sowing before winter. Consequently, autumn and early winter fluxes during WOSR cropping were neglected for calculation of annual N_2O emissions.

The FRE was derived by fitting the model described by Walter et al. (2015), which is based on the methodology in Stehfest and Bouwman (2006), after including the data from this study in their dataset (12 sites from the global meta-analysis of Walter et al. (2015) and five sites from this study with three measurement years and up to five fertilization rates per site). Briefly, a linear mixed effects model (R package *lme4* version 1.1–12, Bates et al. (2015), R package *lmerTest* version 2.0-33, Kuznetsova et al. (2016)) relating $\log_{10}$-transformed annual N_2O fluxes to fertilizer N input was fitted. The model included random intercepts

for the site and year effects and a random slope for the year effect. The log_{10}-transformation accounts for the typical higher heterogeneity of larger N_2O fluxes. As a result, the modeled relationship between N_2O fluxes and fertilization rate is exponential and can be compared to previous models by Walter et al. (2015) and Stehfest and Bouwman (2006). Since the model is nonlinear, emission factors depend on the amount of N fertilizer. We report the emission factor for an N fertilization rate of 200 kg N ha^{-1} as this is the amount beyond which no further yield increases are expected under best management practices (Maidl and Limbrunner, 2008). This is also approximately the recommended and typical fertilization rate of WOSR production in Germany. Following the methodology employed by the JRC (Edwards et al., 2016) it was calculated as:

$$EF = \frac{E_{200} - E_0}{200} \quad (1)$$

where E_{200} and E_0 are emissions (kg N_2O-N ha^{-1} a^{-1}) predicted by the model at 200 kg N and 0 kg N fertilization rate, respectively. Compared to the fitted exponential relationship, this linear emission factor approach slightly overestimates emissions from lower N fertilization and underestimates emissions from higher N fertilization. However, these deviations are small as long as the amount of N fertilizer applied does not differ too substantially from typical fertilization rates. In contrast to the IPCC emission factor (EF1) but in accordance with Stehfest and Bouwman (2006), the emission factor takes into account emissions from crop residues indirectly since crop residue N was not included as N input in the model.

2.4.3. Water-filled pore space

Water-filled pore space (WFPS) was calculated as described by Ruser et al. (1998) using the bulk density measured in the top soil of the study sites and assuming a particle density for the soil of 2.65 g cm^{-3}.

2.4.4. N surplus and oil yield-related N_2O emissions

N surplus was calculated by subtraction of N removed from the field by harvest (dry matter concentration of WOSR seed yield multiplied by N concentration of the seeds) from the respective N fertilizer amount. Oil yield–related N_2O emissions for the respective fertilization treatment were calculated by relating annual N_2O emissions to the amount of oil yield, which was the product of WOSR seed yield and oil concentration in the seeds. For the study site Berge, removal of the straw was also taken into consideration.

A linear mixed effects model of log_{10}-transformed oil yield–related N_2O emissions was used to investigate differences between years and N surplus. Year was included as a fixed effect and site as a random intercept.

Finally, total GHG savings of biodiesel produced from the 180 kg N ha^{-1} WOSR treatment were calculated using the Biograce-I (version 4d, www.biograce.net) excel tool.

2.4.5. Statistical analysis

For each site and for each year we separately ran a Kruskal Wallis One Way Anova on Ranks to detect differences between the treatments concerning oil yield-related emissions. Significant differences were determined using a pairwise multiple comparison procedure (Student-Newman-Keuls, $p < 0.05$).

We calculated simple Spearman Rank Order Correlation Coefficients to test for a relationship between the N_2O and CO_2 release in the postharvest period.

All other statistical analyses were conducted using the R language and environment for statistical computing (version 3.3.2, R core, 2016). Mixed-effects models were fitted using package lme4. Confidence intervals of parameters were estimated using parametric bootstrap. Parameter *p*-values were derived using Satterthwaite's approximation for degrees of freedom (Satterthwaite, 1946, R package lmerTest).

Relationships between nitrous oxide fluxes and explaining variables were investigated using Generalized Additive Models (GAM, R package mgcv version 1.8–16, Wood, 2011), which can model non-linear relationships such as the optimum curve typically observed for N_2O emissions vs. soil moisture.

3. Results and discussion

3.1. Meteorological conditions and seasonal N_2O fluxes

3.1.1. Meteorological conditions

Compared to the long-term mean air temperature at every single site, the annual temperature was higher in all experimental years and, except for Hohenschulen, at all study sites (Table 1). Highest annual air temperatures were measured at all sites in the second year of our investigations (2014), followed by the third year (2015). Additionally, considering annual precipitation, which was, depending on site, lowest in the second or third year of measurements, it became obvious that the climatic conditions during our experiment covered a year representing or slightly exceeding long-term conditions (2013), a year with average precipitation and higher temperatures and one year with rather drier and warmer conditions.

3.1.2. N_2O fluxes during the growing season

Spatial and temporal variability of N_2O fluxes was very high (Fig. 1). At all sites, increases of the N_2O fluxes were often detected after N fertilization in conjunction with rainfall events. The highest N_2O flux rate (670 µg N_2O-N m^{-2} h^{-1}) during the whole investigation period in the treatment with 180 kg N ha^{-1} a^{-1} was measured at the Merbitz study site following a heavy rainfall event (42 mm d^{-1}) one week after the second N fertilization in mid May 2013. Prior to this high N_2O pulse, precipitation after two weeks without rainfall had stimulated N_2O release after the first N fertilizer application at the same site.

Although the magnitude of N_2O flux rates differed between study sites and years, we frequently observed short-term N_2O pulses at all study sites after heavy rainfall events throughout the whole growing season as well as after rewetting of solidly dried soil in summer (Fig. 1).

Similar patterns of the N_2O release from arable soils with increased flux rates after N fertilization and rainfall were often reported and explained with enhanced denitrification due to (i) an increased availability of nitrate as substrate for N_2O production, and (ii) due to the formation of anaerobic conditions as a result of lower gas diffusivity in soil water and thus of a reduced O_2 diffusion into the soil combined with O_2 consumption by soil microbes (Flessa et al., 1995; Corre et al., 1996; MacKenzie et al., 1997).

During the first six weeks postharvest we also frequently observed increased N_2O fluxes following rainfall. Monthly fluxes following harvest were often comparable or even exceeding fluxes after N fertilization (Fig. S1). In the post-harvest period we could not see any differentiating effect on the N_2O flux rates, independent of whether the WOSR residues were incorporated into the soil or remained on the surface. Although we did not include WOSR residue incorporation or surface application in our experimental design, we had study sites where we measured increased N_2O fluxes after rainfall when the residues remained on the surface and also after later incorporation (i.e. Dedelow 2014).

Nett et al. (2015) compared the effect of the incorporation of N-rich cauliflower residues on N_2O fluxes with a treatment where the residues remained as mulch on the soil surface. They did not find significant differences of the N_2O release between these two treatments. Baggs et al. (2003) and Escobar et al. (2010) even reported higher fluxes when legume crop residues remained on the soil surface instead of being incorporated.

In this postharvest period after WOSR, nitrate contents in the top soil were generally elevated (Fig. S2). Mosier et al. (1983) reported a threshold of 10 mg nitrate N kg^{-1} soil above which denitrification rates were independent of the soil nitrate concentration. Nitrate-N

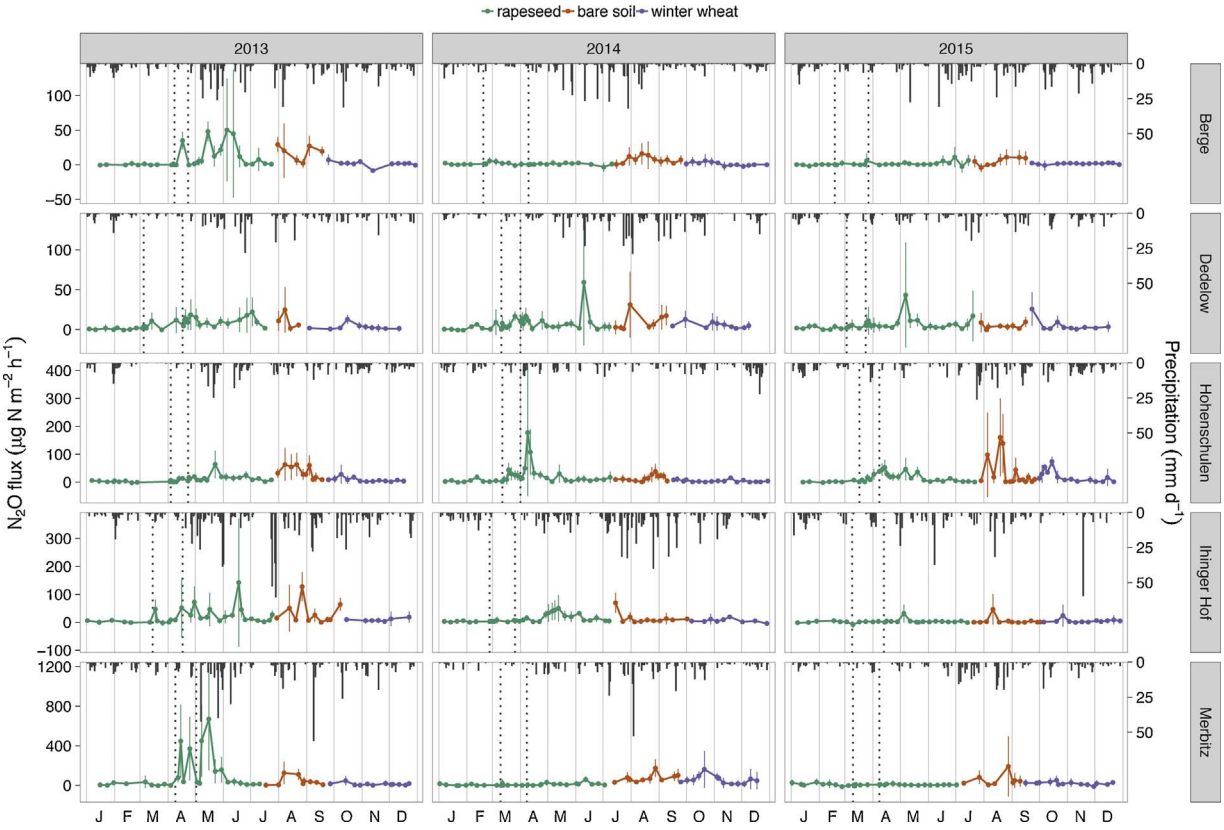

Fig. 1. Mean N_2O flux rate (n = 4) in the treatment with 180 kg N ha^{-1} a^{-1} (colored lines) and daily precipitation (gray bars) as affected by study site and experimental year. Dotted lines represent N fertilization (90 kg N ha^{-1} a^{-1}) each. Note: different y-axis scaling.

concentrations in our study often reached this level immediately after harvest. We therefore concluded that soil nitrate was not a limiting factor for the denitrifying community after harvest and consequently we assume that C availability and corresponding microbial activity and oxygen consumption played a major role as the driver for postharvest N_2O release. This assumption was supported by the statistically highly significant and positive correlations between the N_2O and CO_2 flux rates in the time between harvest of WOSR and seeding of the succeeding winter wheat (Spearman rank correlation coefficients calculated for each study site, all p values were < 0.001, data not shown). It was further supported by the lowest postharvest N_2O flux at the study site Berge, the site with removal of the WOSR straw.

In case of the surface located WOSR residues this C must have been leached from the crop residues during rainfall into the soil or solubilized in regions of the residues with direct soil contact. An alternative explanation could be N_2O production directly from the decomposing plant material (Flessa et al., 2002). Müller et al. (2003) compared the turnover dynamics of different plant residues for modeling purposes. They measured a very low amount of water soluble C in rapeseed straw (~4% of the total C). However, using their algorithm suggested for the relationship between easily available C and the C-to-N ratio we calculated that 36% of the total C in the residues of our treatment with 180 kg N ha^{-1} a^{-1} belonged to the easily decomposable pool.

In contrast to earlier investigations on N_2O fluxes from arable fields or grassland in Germany (Flessa et al., 1995; Kammann et al., 1998; Röver et al., 1998; Kaiser and Ruser, 2000; Ruser et al., 2001), we did not observe considerable N_2O pulses during thawing of frozen soil. We assume that the mild winters in all three experimental years without any severe frost periods were probably the main reason for the low N_2O flux rates observed in our study. It was often shown that frost/thaw induced N_2O pulses increase with increasing duration of frost periods and with severity of the soil freezing (Teepe et al., 2004; Wagner-Riddle et al., 2007; Risk et al., 2013; Xu et al., 2016). Except for the first two weeks of our measurements in 2013, soil temperatures in 10 cm depth did not drop below −2 °C for more than one week at all sites (not shown) and hence the conditions during our field experiment did not enable distinct frost/thaw induced N_2O pulses.

Over the whole data set, we could explain 27% of the variability of the N_2O flux rates (in the treatment with 180 kg N ha^{-1} a^{-1}) with the generalized additive model (Table S1). All smooth terms with a statistically significant contribution to the model results were recorded at the study sites Merbitz, Ihinger Hof, and Hohenschulen, whereas we did not find significant correlations between the smooth terms and the log_{10}-transformed N_2O flux rates at Berge and Dedelow (Fig. S3). We found a relationship between the temperature and soil moisture and the N_2O flux rates for the sites Merbitz, Ihinger Hof, and Hohenschulen. The moisture optimum appeared to be around 50% WFPS. The fluxes at Merbitz were also related to the nitrate contents of the top soil (p < 0.001). Enhanced N_2O flux rates with increasing soil moisture and partly with increasing nitrate contents suggest denitrification as a major source for the N_2O released at the sites Merbitz, Hohenschulen, and Ihinger Hof.

3.2. *Effect of N fertilization on the N_2O fluxes*

Over all sites combined, increasing N fertilization significantly enhanced N_2O flux rates (p < 0.001, Table 3). This effect was more apparent at sites with higher N_2O flux level (Table 2). In contrast, N fertilization effects did not appear at Berge and Dedelow, the sites with the lowest flux levels. Following N fertilization, nitrate and ammonium contents generally increased with increasing N amounts. The increased nitrate contents after N fertilization served as available substrate for N_2O production under conditions supporting denitrification. In contrast, N_2O flux rates were negligible under conditions favorable for nitrification (high ammonium concentrations and soil moisture contents below field capacity).

Table 3
Best linear mixed effects model for rapeseed-specific annual N_2O emissions (N_2O_{annual} in kg N ha^{-1} a^{-1}) for the data in this study pooled with the data from Walter et al. (2015); the variance explained by the random effects, the residual variance and R^2 values are given. R^2 values describe correlation between annual fluxes and predicted (from the fixed effects only and from the complete model) values on the natural scale.

Log_{10} (flux mean) = a + b N_{amount}				
Back transformed to natural scale Flux mean = 10^a $10^{(b \times Namount)}$		Coefficients and 95% confidence intervals in brackets	a = −0.169 (−0.406, −0.068) b = 0.00222 (0.0011, 0.0033)	
Random effects			R^2	
On intercept (variance)	On slope (variance)	Residual variance	fixed effects only	complete model
Site, year (0.100, 0.063)	Year 2e-6	0.021	0.05	0.88

Several field experiments showed positive correlations between the N_2O flux rates and soil nitrate contents (Ruser et al., 2001; Sehy et al., 2003; Jones et al., 2007). Especially for agricultural soils, it was also reported that increased nitrate availability can inhibit the N_2O reductase activity due to the competitive effect of nitrate and N_2O as terminal electron acceptors during denitrification thus stimulating N_2O release from denitrification (Cho and Sakdinan, 1978; Blackmer and Bremner, 1978).

3.3. Effect of study site on the N_2O fluxes

The study site had a significant and strong effect on the N_2O flux rates ($p < 0.001$, Table 3). The median N_2O flux rate over the entire three experimental years in the treatment with a N fertilizer amount of 180 kg N ha^{-1} a^{-1} decreased in the order Merbitz (10.4 µg N_2O-N m^{-2} h^{-1}) > Hohenschulen (7.3 µg N) > Ihinger Hof (5.3 µg N) > Dedelow (2.9 µg N) > Berge (1.1 µg N) (Table 2).

Merbitz, the site with the highest clay content, exhibited the highest flux rates, particularly in the first experimental year (Fig. 1, Table 2). At the sites at Berge and Dedelow, we generally measured low N_2O flux rates. Except for Dedelow in July 2014, N_2O fluxes at these two sites did not exceed 25 µg N_2O-N m^{-2} h^{-1} during the entire experimental period (Fig. 1). These low fluxes were probably a result of the sandy soil texture (Table 1) and the corresponding low water holding capacity enabling good soil aeration. These results are in agreement with N_2O flux measurements in wheat fields on soils differing strongly in soil texture. Pelster et al. (2012) reported annual N_2O emissions between 0.6 and 0.7 kg N_2O-N ha^{-1} from a sandy soil under wheat fertilized with calcareous ammonium nitrate and between 5.1 and 8.3 kg N_2O-N ha^{-1} from a silty clay soil under the same management. In a lysimeter study with three differently textured soils (clay loam, loam, sand) Jamali et al. (2016) reported decreasing N_2O fluxes with increasing portions of sand. They attributed low N_2O fluxes from sandy soils to a lower N_2O production from denitrification, since a higher proportion of macro-pores responsible for drainage and aeration in sandy soils reduces the frequency of conditions favoring denitrification.

Although soil texture at the site Hohenschulen is also sand-dominated (Table 1), N_2O fluxes measured there were distinctively higher than at the sandy sites at Dedelow and Berge (Table 2). Especially for sandy soils, Pelster et al. (2012) assumed a temporal C limitation for denitrifying microorganisms requiring C compounds as electron donator. An increase in the C_{org} content of the soil therefore also enhances the availability of C for the denitrifying microbial community. Stehfest and Bouwman (2006) in their analysis of N_2O emission data from 1008 agricultural soils confirmed the effect of rising N_2O emissions with increasing C_{org} content of topsoil.

The C_{org} content of the topsoil in Hohenschulen was 1.6 and 2.5 times higher than in Berge and Dedelow, respectively (Table 1). For the period between 1st January and the sowing of wheat in autumn of the first experimental year, our flux chambers covered bare soil. The CO_2 flux rates (which are only a rough estimate due to the chamber closing times being optimized for N_2O flux measurements) are therefore an indicator for C mineralization. The mean CO_2 flux rate at Hohenschulen in that period was 86.9 mg CO_2-C m^{-2} h^{-1}. It was 1.6 and 2.3 times higher than the corresponding mean flux rates at Berge and Dedelow. We therefore presume that the higher soil respiration rates at Hohenschulen decreased O_2 availability, thus favoring anaerobic conditions and N_2O production during denitrification whereas O_2 supply in the soils in Berge and Dedelow was sufficient to impede nitrate reduction. This would explain the higher N_2O fluxes compared to the other sandy sites (Table 2).

3.4. Inter-annual variability of the N_2O fluxes

The median annual N_2O flux at all sites exhibited a high variability and the effect of the experimental years was statistically significant and of the same order of magnitude as the site effect (Table 3). The highest inter-annual variability was measured at the study site Ihinger Hof in the treatment with 180 kg N ha^{-1} a^{-1}. At this site, the flux was 5 times higher in 2013 than in 2015 (Table 2).

A main driver for the inter-annual variability of the annual N_2O flux rates was rainfall shortly after N fertilization or harvest. In the treatment with 180 kg N ha^{-1} a^{-1}, the annual N_2O flux rate decreased at all experimental sites in the same order as the annual precipitation decreased (Fig. 2). This clearly indicates that local weather conditions and in particular the amount of rainfall (especially in months with increased mineral N availability as, for example after N fertilization; Fig. S2), play a key role in determining the magnitude of N_2O flux rates.

A high inter-annual variability of N_2O fluxes has often been reported in field studies with N_2O measurements (Dobbie et al., 1999; Pfab et al., 2011; Reeves and Wang, 2015) as well as in modeling approaches from sites with different climate conditions (Leip et al., 2011) or with different climate scenarios (Ben Aoun et al., 2016). Despite a uniform management (N fertilization, crop type) annual N_2O emission varied by up to factor seven between the single experimental years. These differences also resulted from different weather conditions during the study, with rainfall being one of the dominant drivers for N_2O release from soils (Smith et al., 1998; Dobbie et al., 1999; Laville et al., 2011).

3.5. Cumulative N_2O emissions and fertilizer-related N_2O emissions

Due to high variability of N_2O fluxes, cumulative N_2O emissions were also scattered widely over the study sites and experimental years. For the treatment fertilized with 180 kg N ha^{-1} a^{-1}, annual N_2O emission varied between 0.24 kg N_2O-N ha^{-1} a^{-1} (Berge, 2014) and 5.48 kg N_2O-N ha^{-1} a^{-1} (Merbitz, 2013) (Fig. 3).

The magnitude of annual N_2O emissions was in the same range as those assembled by Walter et al. (2015) in their meta-analysis on the effect of N fertilization on N_2O emissions from WOSR fields. For N fertilizer amounts approximately in the same range as our 180 kg N ha^{-1} a^{-1} treatment Walter et al. (2015) reported annual N_2O emissions from WOSR fields ranging between 0.31 and 5.61 kg N_2O-N ha^{-1} a^{-1}.

Since N_2O flux rates were stimulated with increasing N fertilizer amount, cumulative N_2O emissions also increased with N fertilization (Fig. 3). We supplemented the dataset of Walter et al. (2015) with our data, thereby doubling the number of data points, and following their methodology, derived an exponential model relating N_2O emissions to N fertilization (Table 3). The model confirmed a strong impact of study sites and years on annual N_2O fluxes. A nonlinear response of N_2O emissions to N fertilization has often been reported and explained either with an increased N supply strongly exceeding N demand of the crop or with extended periods of increased mineral N supply for N_2O

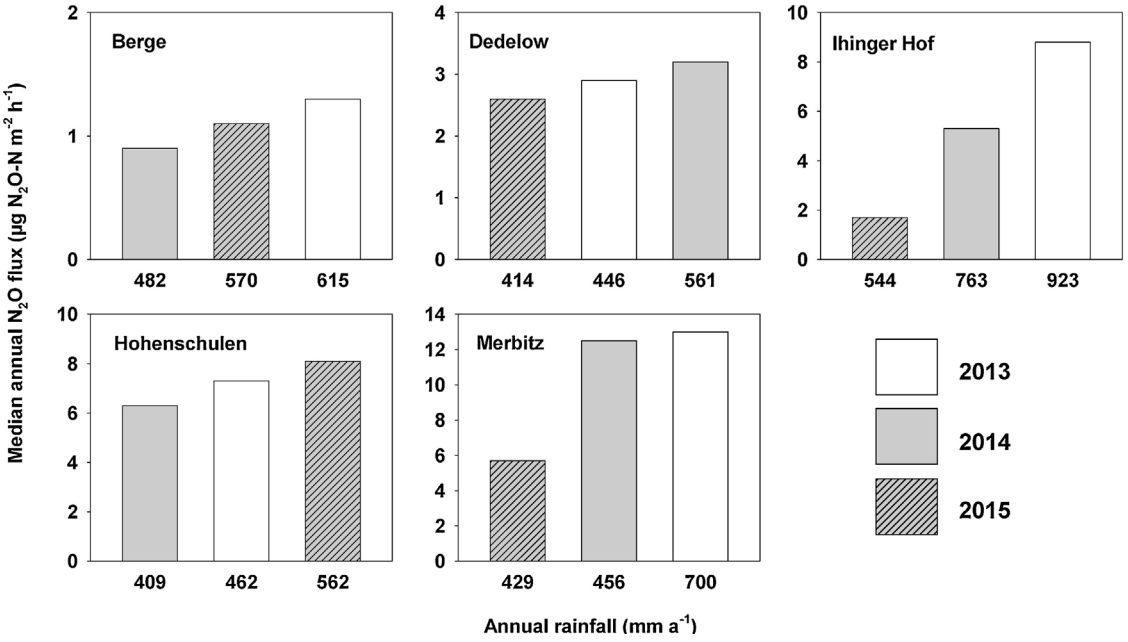

Fig. 2. Median N_2O flux and annual precipitation at the five study sites. Note: different y-axis scaling and non-equidistant x-axis units.

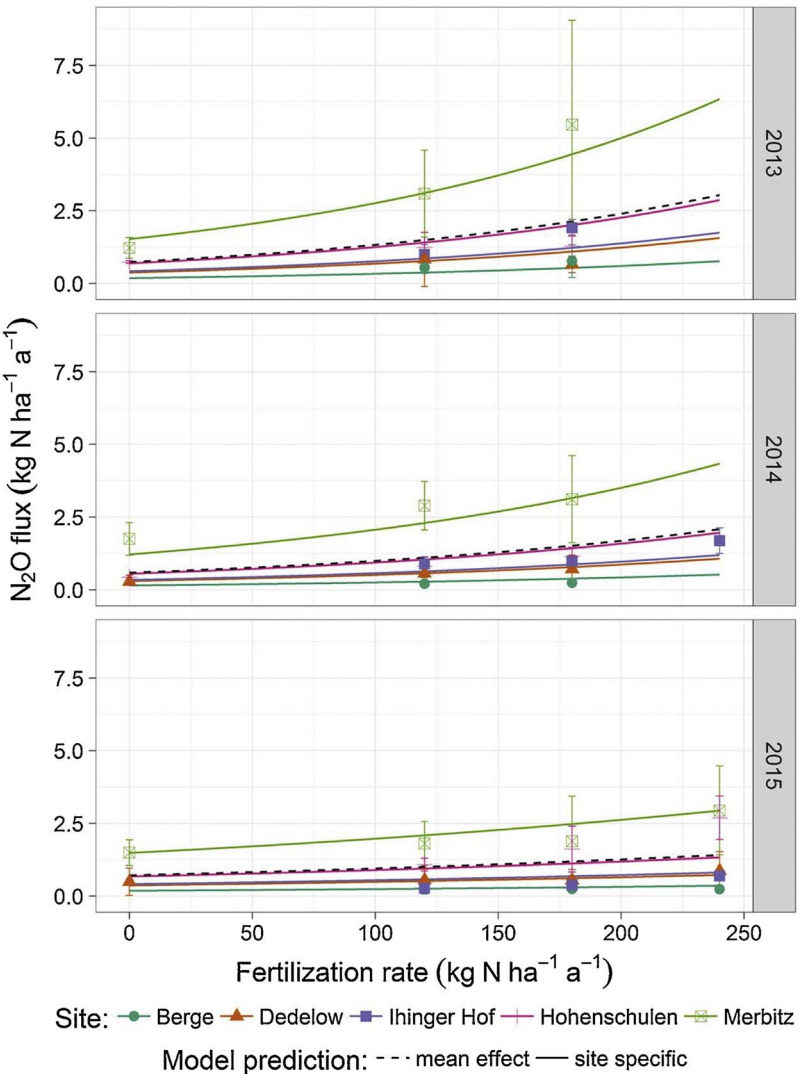

Fig. 3. Mean measured cumulative annual N_2O emission (n = 4, ± standard deviation) at different nitrogen fertilization rates. Lines depict site-specific and mean N_2O emissions modeled with the mixed effects model described in Table 3.

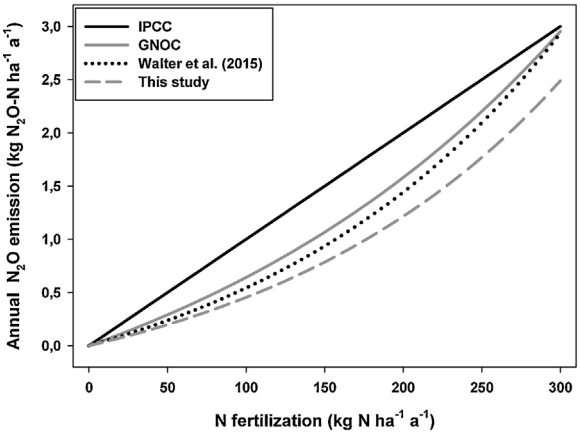

Fig. 4. Effect of different fertilizer-related N_2O emission models on the calculative fertilizer-derived N_2O emission from WOSR fields as affected by N fertilizer amount. IPCC assumes a linear increase of N_2O emission with N application rate (constant emission factor of 1%), GNOC (Edwards et al., 2016), Walter et al. (2015) and the model derived in this study describe fertilizer-derived N_2O emissions as an exponential function of fertilizer N input. The GNOC line represents rapeseed at temperate oceanic sites with 1–3% SOC, pH 5.5–7.3, and medium soil texture.

Table 4
Mean oil yield [Mg ha^{-1} a^{-1}] as affected by study site, N fertilization and experimental year. Different letters indicate statistically significant differences between the N-fertilizer treatments within one year and one study site (Student-Newman-Keuls Method, $p < 0.05$).

Study site	Year	N fertilization				
		kg N ha^{-1} a^{-1}				
		0	60	120	180	240
Berge	2013	1.17 ^{b}	1.29 ^{a,b}	1.38 ^{a,b}	1.38 ^{a,b}	1.72 ^{a}
	2014	1.63 ^{c}	1.92 ^{b,c}	2.28 ^{a,b}	2.42 ^{a}	2.56 ^{a}
	2015	0.97 ^{c}	1.50 ^{b}	1.74 ^{a}	1.87 ^{a}	1.85 ^{a}
	Mean	1.26	1.57	1.80	1.89	2.04
Dedelow	2013	2.36 ^{b}	2.62 ^{a}	2.73 ^{a}	2.79 ^{a}	2.78 ^{a}
	2014	2.43 ^{a}	2.56 ^{a}	2.67 ^{a}	2.74 ^{a}	2.71 ^{a}
	2015	1.97 ^{a}	2.09 ^{a}	2.10 ^{a}	2.05 ^{a}	2.13 ^{a}
	Mean	2.25	2.42	2.50	2.53	2.54
Ihinger Hof	2013	1.59 ^{b}	1.78 ^{a}	2.01 ^{a}	1.96 ^{a}	1.92 ^{a}
	2014	1.22 ^{c}	1.52 ^{b}	1.76 ^{a,b}	1.83 ^{a}	1.91 ^{a}
	2015	1.55 ^{c}	1.64 ^{b,c}	1.76 ^{a,b}	1.86 ^{a}	1.88 ^{a}
	Mean	1.45	1.65	1.84	1.88	1.90
Hohenschulen	2013	1.64 ^{b}	2.01 ^{a}	2.25 ^{a}	2.32 ^{a}	2.40 ^{a}
	2014	2.02 ^{a}	2.35 ^{a}	2.50 ^{a}	2.55 ^{a}	2.60 ^{a}
	2015	1.78 ^{c}	2.04 ^{a,b}	2.21 ^{a}	1.99 ^{b}	2.15 ^{a,b}
	Mean	1.81	2.13	2.32	2.29	2.38
Merbitz	2013	1.61 ^{b}	1.75 ^{a,b}	1.87 ^{a,b}	1.88 ^{a,b}	1.96 ^{a}
	2014	1.18 ^{d}	1.61 ^{c}	2.09 ^{b}	2.23 ^{b}	2.38 ^{a}
	2015	1.11 ^{b}	1.30 ^{b}	1.67 ^{a}	1.78 ^{a}	1.64 ^{a}
	Mean	1.30	1.55	1.88	1.96	2.00

production (van Groeningen et al., 2010; Hoben et al., 2011; Shcherbak et al., 2014).

The German legislation on N fertilization (DüV, 2006) currently allows for an N surplus (N fertilization minus N removal with harvest) of 60 kg N ha^{-1} a^{-1} (mean value of three years). Assuming a high WOSR yield of 5 Mg ha^{-1} a^{-1} we can deduce a crop demand of 227 kg N ha^{-1} a^{-1} (5 Mg ha^{-1} a^{-1} × 45.4 kg N Mg^{-1}, cf. Table 1 of DüV, 2006). On a legislative basis, it can therefore be expected, that N fertilization in German WOSR production potentially varies between 0 and 287 kg N ha^{-1}. Over this range of N fertilization, all proposed nonlinear models for the N_2O emission – N fertilization rate relationship (Fig. 4) result in lower fertilizer-related N_2O emissions when compared to the linear IPCC Tier 1 approach.

Comparing the impact of different emission factors for direct N_2O field emissions from WOSR cultivation in Poland, Syp et al. (2016) also reported higher N_2O emission calculated with the BioGrace approach (IPCC default values, Tier 1) compared to the GNOC.

Assuming a fertilizer amount of 200 kg N ha^{-1} a^{-1} the global FRE factor derived from the exponential model was 0.6% (CI: 0.31%–1.00%). This factor is within the uncertainty range of the EF1 IPCC emission factor (0.3%–3%), but about 40% lower than the IPCC default value and was also lower than the FRE calculated by GNOC and by Walter et al. (2015) (Fig. 4).

One reason for the lower FRE in our experiment may be the fact that two of our five study sites have sandy, well aerated soils with low C_{org} contents. These were chosen because they are representative for a large part of the German WOSR production area.

A second reason might be the absence of distinct frost/thaw cycles at all study sites. As a consequence, the absence of frost/thaw cycles results in N_2O emissions about half as high as in case of frost/thaw cycle occurrence. However, such mild winters with less frost/thaw cycles seem to have become more frequent in Germany as a result of ongoing climate change (Kreyling and Henry, 2011).

The low rapeseed FRE factor is in good agreement with results from recent studies in the UK which also observed lower FRE factors than the IPCC default (Bell et al., 2015). Based on multiple field experiments with different crops including WOSR a new national emission factor of 0.46% was derived for the UK (Sylvester-Bradley et al., 2015). Similarly, a recent multi-site study in France, again including WOSR in the field experiments, observed lower N_2O emissions than expected from the IPCC default emission factor (Le Gall et al., 2014).

3.6. Oil yield and yield-related N_2O emissions

The highest mean seed yields over all three experimental years were achieved in Dedelow (5.38 Mg ha^{-1} a^{-1}) and in Hohenschulen (4.67 Mg ha^{-1} a^{-1}). At the remaining three sites (Berge, Merbitz and Ihinger Hof) seed yield was lower and ranged between 3.97 and 4.22 Mg ha^{-1} a^{-1}. The mean seed yields over all sites were 4.54, 4.90,

Table 5
Mean oil yield-related N_2O emission [kg N_2O-N Mg^{-1} oil ha^{-1}] as affected by study site, N fertilization and experimental year. Different letters indicate statistically significant differences between the N fertilizer treatments within one year and one study site (Student-Newman-Keuls Method used for comparison of three or more treatments, *t*-test for comparison of two treatments, $p < 0.05$).

Study site	Year	N fertilization			
		kg N ha^{-1} a^{-1}			
		0	120	180	240
Berge	2013	–	0.50 ^{a}	0.58 ^{a}	–
	2014	–	0.07 ^{a}	0.10 ^{a}	–
	2015	–	0.31 ^{a}	0.15 ^{a}	0.14 ^{a}
	Mean$^{\$}$	–	0.29	0.28	–
Dedelow	2013	–	0.18 ^{a}	0.24 ^{a}	–
	2014	0.13^{a}	0.23 ^{a}	0.26 ^{a}	–
	2015	0.25^{a}	0.27 ^{a}	0.30 ^{a}	0.35 ^{a}
	Mean$^{\$}$	–	0.23	0.27	–
Ihinger Hof	2013	–	0.53 ^{b}	0.97 ^{a}	–
	2014	–	0.56 ^{a}	0.52 ^{a}	0.99 ^{a}
	2015	–	0.19 ^{b}	0.19 ^{b}	0.41 ^{a}
	Mean$^{\$}$	–	0.42	0.56	–
Hohenschulen	2013	0.51 ^{a}	0.58 ^{a}	0.55 ^{a}	–
	2014	0.21 ^{b}	0.31 ^{b}	0.47 ^{a}	–
	2015	–	0.54 ^{b}	0.85 ^{a,b}	1.52 ^{a}
	Mean$^{\$}$	–	0.49	0.56	–
Merbitz	2013	0.75^{a}	1.59 ^{a}	2.98 ^{a}	–
	2014	1.48 ^{a}	1.45 ^{a}	1.36 ^{a}	–
	2015	1.43^{a}	1.03 ^{a}	1.12 ^{a}	1.93 ^{a}
	Mean$^{\$}$	1.22	1.36	1.82	–

– not determined/not calculated.
$^{\$}$ Mean values were only calculated for treatments with 3 years measurements.

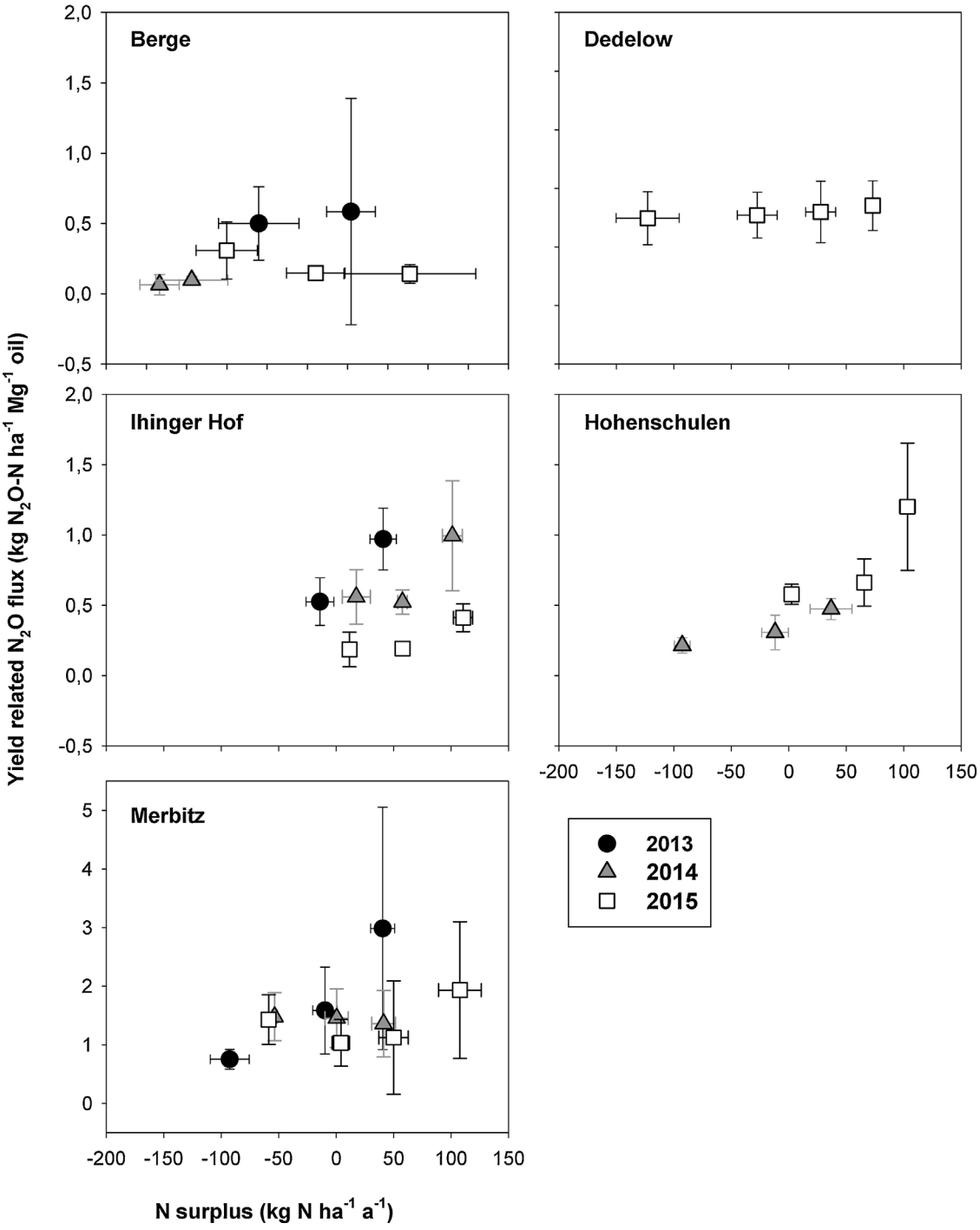

Fig. 5. Relationship between mean N surplus and mean oil yield-related N_2O emission (n = 4, ± standard deviation) as affected by study site and experimental year. At the site Berge straw removal was also taken into consideration for the calculation of the N surplus. Note: different y-axis scaling.

and 4.03 Mg ha^{-1} a^{-1} in 2013, 2014, and 2015, respectively. These yields follow the pattern of mean German WOSR yields, which were 3.96, 4.48, and 3.91 Mg ha^{-1} a^{-1} in 2013, 2014, and 2015, respectively (German Federal Statistical Office, 2017). Research studies commonly achieve better yields than commercial farms, but the identical annual pattern emphasizes the representativeness of our compilation of study sites for German WOSR production.

The reason for the high yield at Dedelow remains unclear. A possible explanation is a pool of easily mineralizable N, resulting from long-term application of organic fertilizers to silage maize and sugar beet approximately every second year before our experiment. However, C_{org} and total N contents at this site were comparatively low. Unfortunately, we did not determine N contents of the WOSR seeds in every year and at every site since we focused on oil yield as the relevant target yield. However, for the year 2015 we determined an N uptake in the seeds of 123 kg N ha^{-1} in the unfertilized treatment at Dedelow. The only study site with N uptake measurements in the unfertilized treatment in the same year was Merbitz with 59 kg N ha^{-1}. Further N uptake data for unfertilized treatments at other sites than Dedelow in 2013 and 2014 varied between 54 and 93 kg N ha^{-1}. The high N uptake in the rapeseeds at Dedelow was not only a result of increased biomass growth but also enhanced N concentrations (not shown). Both, high biomass and N concentration in the seed of the unfertilized treatment in Dedelow indicates a high and easily available N delivery at

Table 6
GHG emissions and savings of biofuel produced from the 180 kg N ha^{-1} a^{-1} treatment according to BioGrace-I GHG calculation tool (version 4d). Standard values were used for all values not given in the Table.

Mineral N fertilizer	kg N ha^{-1} a^{-1}	180	180	180	180	180	180
Production of N fertilizer	g CO_2-eq kg^{-1}	3652[1]	3652[1]	3652[1]	3652[1]	3652[1]	3652[1]
Emission factor for direct N_2O field emissions from mineral fertilizer N	%	1	1	0.6	0.6	0.6	0.6
Emission factor for direct N_2O field emissions from crop residues (IPCC default)	%	1	1	1	1	0.6	0.6
Fresh matter seed yield	Mg ha^{-1} a^{-1}	4.99	4.99	4.99	4.99	4.99	4.99
Soil water saturation high?		yes[2]	yes[2]	yes[2]	yes[2]	yes[2]	yes[2]
Direct and indirect N_2O field emissions	kg N_2O ha^{-1} a^{-1}	5.87	5.87	4.77	4.77	4.09	4.09
	kg N_2O-N ha^{-1} a^{-1}	3.74	3.74	3.04	3.04	2.60	2.60
Fossil fuel reference	g CO_2-eq MJ^{-1}	83.8	95.1[3]	83.8	95.1[3]	83.8	95.1[3]
GHG emission reduction vs reference	%	44	51	47	54	50	56

[1] Biograce standard emission factor for CAN.
[2] This setting accounts for 30% loss of input N via nitrate leaching. The alternative is zero nitrate leaching.
[3] New reference value (fossil diesel) according to EU directive 2015/1513 (EU, 2015).

this site.

The median flux over all sites and years in the treatment with 180 kg N ha^{-1} a^{-1} related to grain yield was 0.22 kg N_2O-N Mg^{-1} oilseed grain.

Oil yield varied between 0.97 and 2.79 Mg ha^{-1} a^{-1} (Table 4), the median oil yield was 2.00 Mg ha^{-1} a^{-1}. Due to the high seed yield, oil yield was also highest at study site Dedelow, where 2.25 Mg ha^{-1} a^{-1} was achieved even in the unfertilized treatment. With few exceptions (Hohenschulen 2015 and Merbitz 2014) N fertilization higher than 120 kg N ha^{-1} did not result in statistically significant increases in oil yield.

This result is in agreement with Hegewald et al. (2016) who reported only small increases in oil yield (0.04 Mg ha^{-1} a^{-1}) when N fertilization was increased from 120 to 180 kg N ha^{-1} a^{-1} in a study on the effect of different preceding crops on WOSR yield. As mentioned by Rathke et al. (2006) N fertilization increases the crude protein content of rapeseeds at the expense of oil concentration. We also observed declining oil contents with increasing N fertilization, however, due to higher dry matter development with increasing N supply, oil yields were stable over the N fertilizer range between 120 and 240 kg N ha^{-1} a^{-1} (Table 4).

Oil yield–related N_2O emissions varied depending on site and year. The median yield-related N_2O emission over the entire data set was 0.46 kg N_2O-N Mg^{-1} oil. Cumulative annual N_2O emissions, and consequently oil yield-related N_2O emissions, were lowest at study sites Berge and Dedelow (Table 5). For 2014 and 2015 we found a tendency at Dedelow for increasing yield-related emission with increasing N fertilization.

In contrast, oil-yield related N_2O emissions were distinctly higher in all years for the site Merbitz with its silty Chernozem soil and also higher for the sites Ihinger Hof and Hohenschulen (Table 5). Increasing N fertilization at these three sites by 60 kg N ha^{-1} from 120 to 180 kg N ha^{-1} a^{-1} resulted in a slight increase (approximately 0.15 kg N_2O-N Mg^{-1}) in yield-related N_2O emissions whereas the application of another 60 kg N ha^{-1} a^{-1} (in total 240 kg N ha^{-1} a^{-1}) approximately doubled the yield-related emissions in the respective years with trace gas measurements indicating a threshold for strongly enhanced yield-related N_2O emissions between fertilization intensities of 180 and 240 kg N ha^{-1} a^{-1}.

Different functions have been used to visualize the relation of yield-related N_2O emissions to N surplus. Van Groenigen et al. (2010) used an exponential function to describe the yield-related N_2O emissions with N surplus in silage maize. Walter et al. (2015) fitted a segmented linear function to their WOSR yield dry mass-related N_2O emissions and found a critical N surplus of 80 kg N ha^{-1} where the yield-related N_2O emissions substantially increased. We found neither a clear threshold in our experiment nor a simple global relationship. This can be attributed to the high inter-annual variability of the N_2O emissions (Fig. 5). Especially in 2013, N_2O emissions were driven by fertilization whereas emissions in 2014 and 2015 did depend less on fertilization and were generally lower. Oil yield-related N_2O emissions at the study sites Ihinger Hof and Merbitz increased with a small N surplus in 2013, the year with the highest precipitation, whereas the threshold for increased yield-related N_2O emissions under drier conditions in 2014 and 2015 was approximately +50 kg N ha^{-1} N surplus. The site Hohenschulen also exhibited a distinct increase of yield-related N_2O emission with rising N surplus, whereas the low emission sites Berge and Dedelow did not respond to varying N surpluses. The removal of WOSR straw at the site Berge resulting in lower N surplus values might have additionally affected N_2O emissions.

Calculating the GHG balance of biodiesel produced from the 180 kg N ha^{-1} a^{-1} WOSR treatment according to current EU RED methodology (i.e., using the IPCC emission factor of 1%) resulted in GHG savings of 44% and 51% compared to the current and updated fossil fuel reference, respectively (Table 6). Note that both yield (4990 kg ha^{-1} a^{-1}) and N fertilization of the 180 kg N treatment were much higher than the EU RED default values (3113 kg ha^{-1} a^{-1} with 137.4 kg N fertilization). Substituting the EF1 IPCC emissions factor with the rapeseed specific emissions factor of 0.6% for the calculation of direct N_2O field emissions from fertilizer N input improved GHG savings to 47% and 54%, respectively. If we assume that the same WOSR-specific emission factor is also valid for emissions from the crop residues, GHG savings even achieve 50% and 56%, respectively. Thus, a full update of the emission factor results in a calculative increase of GHG savings by five to six percentage points. As a result the 50% goal of the EU RED would be achieved even with the original reference value.

4. Conclusions

With our study covering three years at five study sites in representative regions of German WOSR production, we doubled the experimental base (now 86 annual flux values) for the deduction of an emission factor for fertilizer-related N_2O emissions from WOSR cropping. Our measurements confirmed the result of Walter et al. (2015) that fertilizer-related N_2O emissions from WOSR are markedly lower than the (linear) one percent default value proposed by the IPCC Tier 1 approach. For a common N fertilizer amount of 200 kg N ha^{-1} a^{-1} in German WOSR cultivation the fertilizer-related emission factor developed from our data set combined with the data assembled by Walter et al. (2015) is 0.6%. Applying this WOSR-specific emission factor to the N fertilizer amount used in our experiment (180 kg N ha^{-1} a^{-1}), which is similar to the amounts used under best agricultural management conditions, we could show, that the GHG reduction criteria stipulated by the Renewable Energy Directive for biofuels can be fulfilled for existing biofuel plants: the GHG emissions from biofuel production achieved more than 50% GHG savings compared to fossil diesel exploration and consumption, especially if best management practices are adopted in WOSR cultivation.

Our oil yield results indicate that there is potential for reduction of N fertilization in comparison to agricultural practice recommendations

without oil yield losses. This would be an excellent mitigation option due to avoiding GHG emissions during fertilizer production and the reduction of direct and indirect N_2O field emissions.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.agee.2017.07.039.

References

Baggs, E.M., Stevenson, M., Pihlatie, M., Regar, A., Cook, H., Cadisch, G., 2003. Nitrous oxide emissions following application of residues and fertilizer under zero and conventional tillage. Plant Soil 254, 361–370.

Bates, D., Maechler, M., Bolker, B., Walker, S., 2015. Fitting linear mixed-effects models using lme4. J. Stat. Softw. 67, 1–48.

Bell, M.N., Hinton, N., Cloy, J.M., Topp, C.F.E., Rees, R.M., Cardenas, L., Scott, T., Webster, C., Ashton, R.W., Whitmore, A.P., Balshaw, H., Paine, F., Goulding, K.W.T., Chadwick, D.R., 2015. Nitrous oxide emissions from UK arable soils Fluxes, emission factors and mitigation. Agric. Ecosyst. Environ. 212, 134–147.

Ben Aoun, W., El Akkari, M., Flénet, F., Jacquet, F., Benoit, G., 2016. Recommended fertilization practices improve the environmental performance of biodiesel from winter oilseed rape in France. J. Clean. Prod. 139, 242–249.

Blackmer, A.M., Bremner, J.M., 1978. Inhibitory effect of nitrate on reduction of N_2O to N_2 by soil microorganisms. Soil Biol. Biochem. 10, 187–191.

Bouwman, A.F., 1996. Direct emission of nitrous oxide from agricultural soils. Nutr. Cycl. Agroecosyst. 46, 53–70.

Bremner, J.M., 1997. Sources of nitrous oxide in soils. Nutr. Cycl. Agroecosyst. 49, 7–16.

Butterbach-Bahl, K., Baggs, E.M., Dannenmann, M., Kiese, R., Zechmeister-Boltenstern, S., 2013. Nitrous oxide emissions from soils: how well do we understand the processes and their controls? Phil. Trans. R. Soc. B: Biol. Sci. 368.

Carré, P., Pouzet, A., 2014. Rapeseed market, worldwide and in Europe. OCL 21, D102.

Cho, C.M., Sakdinan, L., 1978. Mass spectrometric investigation on denitrification. Can. J. Soil Sci. 58, 443–457.

Clais, P., Sabine, C., Bala, G., Bopp, L., Brovkin, V., Canadell, J., Chhabra, A., DeFries, R., Galloway, J., Heimann, M., Jones, C., Le Quéré, C., Myneni, R.B., Piao, S., Thornton, P., 2013. Carbon and other biogeochemical cycles. In: Stocker, T.F., Qin, D., Plattner, G.-K., Tignor, M., Allen, S.K., Boschung, J., Nauels, A., Xia, Y., Bex, V., Midgley, P.M. (Eds.), Climate Change 2013: The Physical Science Basis Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press Cambridge, United Kingdom and New York, NY USA, pp. 465–570.

Corre, M.D., Van Kessel, C., Pennock, D.J., 1996. Landscape and seasonal patterns of nitrous oxide emissions in a semiarid region. Soil Sci. Soc. Am. J. 60, 1806–1815.

Crutzen, P.J., Mosier, A.R., Smith, K.A., Winiwarter, W., 2008. N_2O release from agro-biofuel production negates global warming reduction by replacing fossil fuels. Atmos. Chem. Phys. 8, 389–395.

Crutzen, P.J., 1981. Atmospheric chemical processes of the oxides of nitrogen including nitrous oxide. In: Delwiche, C.C. (Ed.), Denitrification, Nitrification and Atmospheric N_2O. Wiley, Chichester, pp. 17–44.

Dobbie, K.E., McTaggart, I.P., Smith, K.A., 1999. Nitrous oxide emissions from intensive agricultural systems: variations between crops and seasons; key variables; and mean emission factors. J. Geophys. Res. 104, 26891–26900.

Dufossé, K., Gabrielle, B., Drouet, J.-L., Bessou, C., 2013. Using agroecosystem modeling to improve the estimates of N_2O emissions in the life-cycle assessment of biofuels. Waste Biomass Valor. 4, 593–606.

DüV, Verordnung über die Anwendung von Düngemitteln, Bodenhilfsstoffen, Kultursubstraten und Pflanzenhilfsmitteln nach den Grundsätzen der guten fachlichen Praxis beim Düngen (Düngeverordnung – DüV, German legislation on fertilization). Bundesgesetzblatt Jahrgang 2006 Teil I Nr. 2, 2006.

EU, 2015. Directive (EU) 2015/1513 of the European Parliament and of the Council of 9 September 2015 amending Directive 98/70/EC relating to the quality of petrol and diesel fuels and amending Directive 2009/28/EC on the promotion of the use of energy from renewable sources. Off. J. Eur. Union L 239, 1.

Edwards, R., Mulligan, D., Giuntoli, J., Agostini, A., Boulamanti, A., Koeble, R., Marelli, L., et al., 2013. Assessing GHG Default Emissions from Biofuels in EU Legislation. Review of Input Database to Calculate Default GHG Emissions, Following Expert Consultation 22–23 November 2011, Ispra (Italy). JRC Scientific and Policy Reports. EUR 25595 EN. European Commission Joint Research Centre Institute for Energy and Transporthttp://dx.doi.org/10.2788/66442.

Edwards, R., O'Connell, A., Padella, M., Mulligan, D., Giuntoli, J., Agostini, A., Koeble, R., Moro, A., Marelli, L., 2016. Definition of Input Data to Assess GHG Default Emissions from Biofuels in EU Legislation, JRC Science for Policy Report EUR 26853 EN.

Escobar, L.F., Amado, T.J.C., Bayer, C., Chavez, L.F., Zanatta, J.A., Fiorin, J.E., 2010. Postharvest nitrous oxide emissions from a subtropical oxisol as influenced by summer crop residues and their management. Rev. Bras. Cienc. Solo 34, 435–442.

FAO, 2016. http://faostat3. fao.org/download/Q/QC/E. (Accessed 29.09.2016).

Flessa, H., Dörsch, P., Beese, F., 1995. Seasonal variation of N_2O and CH_4 fluxes in differently managed arable soils in southern Germany. J. Geophys. Res. 100, 23115–23124.

Flessa, H., Potthoff, M., Loftfield, N., 2002. Greenhouse estimates of CO_2 and N_2O emissions following surface application of grass mulch: importance of indigenous microflora of mulch. Soil Biol. Biochem. 34, 875–879.

German Federal Statistical Office, 2017, DESTATIS database, www.destatis.de (Accessed 16.02.2017).

Granli, T., Bøckman, O.C., 1994. Nitrous oxide from agriculture. Nor. J. Agric. Sci. (Suppl. 12).

Hamelinck, C., De Loveinfosse, I., Koper, M., et al., 2013. Renewable Energy Progress and Biofuels Sustainability. Ecofys, London.

Hegewald, H., Koblenz, B., Wensch-Dorendorf, M., Christen, O., 2016. Impacts of high intensity crop rotation and N management on oilseed rape productivity in Germany. Crop Pasture Sci. 67, 439–449.

Henke, J., Breustedt, G., Sieling, K., Kage, H., 2007. Impact of uncertainty on the optimum nitrogen fertilization rate and agronomic, ecological and economic factors in an oilseed rape based crop rotation. J. Agric. Sci. 145, 455–468.

Hoben, J.P., Gehl, R.J., Millar, N., Grace, P.R., Robertson, G.P., 2011. Nonlinear nitrous oxide (N_2O) response to nitrogen fertilizer in on-farm corn crops of the US Midwest. Gobal Change Biol. 17, 1140–1152.

Honaker, J., King, G., Blackwell, M., 2011. Amelia II: a program for missing data. J. Stat. Softw. 45, 1–47.

Hong, J., 2012. Uncertainty propangation in life cycle assesment of biodiesel versus diesel: global warming and non-renewable energy. Bioresour. Technol. 113, 3–7.

Huber, P.J., 1981. Robust Statistics. Wiley.

Hutchinson, G.L., Livingston, G.P., 2001. Vents and seals in non-steady-state chambers used for measuring gas exchange between soil and the atmosphere. Eur. J. Soil Sci. 52, 675–682.

Hutchinson, G.L., Mosier, A.R., 1981. Improved soil cover method for field measurement of nitrous oxide fluxes. Soil Sci. Soc. Am. J. 45, 311–316.

IPCC, et al., 2006. IPCC. In: Egglestone (Ed.), Guidelines for National Greenhouse Gas Inventories. Prepared by the National Greenhouse Gas Inventories Programme. IGES, Japan.

IPCC, 2013. Summary for policymakers. In: Stocker, T.F., Qin, D., Plattner, G.-K., Tignor, M., Allen, S.K., Boschung, J., Nauels, A., Xia, Y., Bex, V., Midgley, P.M. (Eds.), Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, United Kingdom and New York, NY USA, pp. 1–27.

IPCC,, 2001. Climate change 2001: the scientific basis. In: Houghton, J.T., Ding, Y., Griggs, D.J., Noguer, M., van der Linden, P.J., Dai, X., Maskell, K., Johnson, C.A. (Eds.), Contribution of Working Group I to the Third Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press Cambridge, United Kingdom and New York, NY USA.

IUSS Working Group, 2015. World Reference Base for Soil Resources 2014, Update 2015. International Soil Classification System for Naming Soils and Creating Legends for Soil Maps. World Soil Resources Reports No. 106. FAO, Rome.

Jamali, H., Quale, W., Scheer, C., Rowlings, D., Baldock, J., 2016. Effect of soil texture and wheat plants on N_2O fluxes: a lysimeter study. Agric. For. Meteorol. 223, 17–29.

Jones, S.K., Rees, R.M., Skiba, U.M., Ball, B.C., 2007. Influence of organic and mineral N fertiliser on N_2O fluxes from a temperate grassland. Agric. Ecosyst. Environ. 121, 74–83.

Jungkunst, H.F., Freibauer, A., Neufeldt, H., Bareth, G., 2006. Nitrous oxide emissions from agricultural land use in Germany— a synthesis of available annual field data. J. Plant Nutr. Soil Sci. 169, 341–351.

Kaiser, E.A., Ruser, R., 2000. Nitrous oxide emissions from arable soils in Germany – an evaluation of six long-term field experiments. J. Plant Nutr. Soil Sci. 163, 249–260.

Kammann, C., Grünhage, L., Müller, C., Jacobi, S., Jäger, H.J., 1998. Seasonal variability and mitigation options for N_2O emissions from differently managed grasslands. Environ. Pollut. 102, 179–186.

Kim, D.-G., Hernandez-Ramirez, G., Giltrap, D., 2013. Linear and nonlinear dependency of direct nitrous oxide emissions on fertilizer nitrogen input: a meta-analysis. Agric. Ecosyst. Environ. 168, 53–65.

Kreyling, J., Henry, H.A.L., 2011. Vanishing winters in Germany: soil frost dynamics and snow cover trends: and ecological implications. Clim. Res. 46, 269–276.

Kuznetsova, A., Bruun Brockhoff, P., Bojesen Christensen, R.H., 2016. LmerTest: Tests in Linear Mixed Effects Models. R Package Version 2. pp. 0–32. https://CRAN.R-project.org/package=lmerTest.

Laville, P., Lehuger, S., Loubet, B., Chaumartin, F., Cellier, P., 2011. Effect of management, climate and soil conditions on N_2O and NO emissions from an arable crop rotation using high temporal resolution measurments. Agric. For. Meteorol. 151, 228–240.

Le Gall, C., Jeuffroy, M.H., Hénault, C., Phyton, Y., Cohan, J.P., Parnaudeau, V., Mary, B., Compere, P., Tristant, D., Duval, R., Cellier, P., 2014. Analyser et estimer les émissions de N_2O dans les systèmes de grande cultures francais. Innov. Agronomiques 34, 97–112.

Müller, T., Magid, J., Jensen, L.S., Nielsen, N.E., 2003. Decomposition of plant residues of

different quality in soil—DAISY model calibration and simulation based on experimental data. Ecol. Model. 166, 3–18.
MacKenzie, A.F., Fan, M.X., Cadrin, F., 1997. Nitrous oxide emission as affected by tillage, corn-soybean-alfalfa and nitrogen fertilization. Can. J. Soil Sci. 77, 145–152.
Maidl, F.-X., Limbrunner, B., 2008. So optimieren Sie Ihre N-Düngung in Raps (Publication in german). Top Agrar 2 (2008), 64–66.
Malagoli, P., Laine, P., Rossato, L., Ourry, A., 2005. Dynamics of nitrogen uptake and mobilization in field grown winter oilseed rape (Brassica napus) from stem extension to harvest. 1. Global N flows between vegetative and reproductive tissues I relation to leaf fall and their residueal. N. Ann. Bot. 95, 853–861.
McSwiney, C.P., Robertson, G.P., 2005. Nonlinear response of N_2O flux to incremental fertilizer addition in a continous maize (Zea mays L.) cropping system. Gobal Change Biol. 11, 1712–1719.
Meier, U., 2001. Growth Stages of Mono- and Dicotyledonous Plants. BBCH Monograph, 2nd ed. Federal Biological Research Centre for Agriculture and Forestry, Braunschweig.
Mosier, A.R., Parton, W.J., Hutchinson, G.L., 1983. Modelling nitrous oxide evolution from cropped and native soils. In: Hallberg, R. (Ed.), Environmental Biogeochemistry. Ecological Bulletin, Stockholm, pp. 229–241.
Nett, L., Fuß, R., Flessa, H., Fink, M., 2015. Emissions of nitrous oxide and ammonia from a sandy soil following surface application and incorporation of cauliflower leaf residues. J. Agric. Sci. 153, 1341–1352.
Pedersen, A.R., Petersen, S.O., Schelde, K., 2010. A comprehensive approach to soil-atmosphere trace-gas flux estimation with static chambers. Eur. J. Soil Sci. 61, 888–902.
Pelster, D.E., Chatigny, M.H., Rochette, P., Angers, D.A., Rieux, C., Vanasse, A., 2012. Nitrous oxide emissions respond differently to mineral and organic nitrogen sources in contrasting soil types. J. Environ. Qual. 41, 427–435.
Pfab, H., Palmer, I., Buegger, F., Fiedler, S., Müller, T., Ruser, R., 2011. N_2O fluxes from a Haplic Luvisol under intensive production of lettuce and cauliflower as affected by different N fertilization strategies. J. Plant Nutr. Soil Sci. 174, 545–553.
R Core Team, 2016. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
Röver, M., Heinemeyer, O., Kaiser, E.A., 1998. Microbial induced nitrous oxide emissions from an arable soil during winter. Soil Biol. Biochem. 14, 1859–1865.
RED, 2009. 28/EC of the European Parliament and of the Council of 23 April 2009 on the promotion of the use of energy from renewable sources and amending and subsequently repealing Directives 2001/77/EC and 2003/30/EC. Off. J. Eur. Union 140, 16–45.
Rathke, G.-W., Behrens, T., Diepenbrock, W., 2006. Integrated nitrogen management strategies to improve seed yield, oil content and nitrogen efficiency of winter oilseed rape (Brassica napus L.): a review. Agric. Ecosyst. Environ. 117, 80–108.
Ravishankara, A.R., Daniel, J.S., Portman, R.W., 2009. Nitrous oxide (N_2O): The dominant ozone-depleting substance emitted in the 21st century. Science 326, 123–125.
Reeves, S., Wang, W., 2015. Optimum sampling time and frequency for measuring N_2O emissions from a rain-fed cereal cropping system. Sci. Total Environ. 530–531, 219–226.
Risk, N., Snider, D., Wagner-Riddle, C., 2013. Mechanisms leading to enhanced soil nitrous oxide fluxes induced by freeze-thaw cycles. Can. J. Soil Sci. 93, 401–414.
Ruser, R., Flessa, H., Schilling, R., Steindl, H., Beese, F., 1998. Soil compaction and fertilization effects on nitrous oxide and methane fluxes in potato fields. Soil Sci. Soc. Am. J. 62, 1587–1595.
Ruser, R., Flessa, H., Schilling, R., Beese, F., Munch, J.C., 2001. Effect of crop-specific field management and N fertilization on N_2O emissions from a fine-loamy soil. Nutr Cycl. Agroecosyst. 59, 177–191.
Satterthwaite, F.E., 1946. An approximate distribution of estimates of variance components. Biomet. Bull. 2, 110–114.
Sehy, U., Ruser, R., Munch, J.C., 2003. Nitrous oxide fluxes from maize fields: relationship to yield, site-specific fertilization, and soil conditions. Agric. Ecosyst. Environ. 99, 97–111.
Shaw, L.J., Nicol, G.W., Smith, Z., Fear, J., Prosser, J.I., Baggs, E.M., 2006. Nitrosospira spp. can produce nitrous oxide via a nitrifier denitrification pathway. Environ. Microbiol. 8, 214–222.
Shcherbak, I., Millar, N., Robertson, G.P., 2014. Global metaanalysis of the nonlinear response of soil nitrous oxide (N_2O) emissions to fertilizer nitrogen. PNAS 111, 9199–9204.
Sieling, K., Kage, H., 2010. Efficient N management using winter oilseed rape. A review. Agron. Sustain. Dev. 30, 271–279.
Smith, K.A., Searchinger, T.D., 2012. Crop-based biofuels and associated environmental concerns. Glob. Change Biol. Bioenergy 4, 479–484.
Smith, K.A., McTaggart, I.P., Dobbie, K.E., Conen, F., 1998. Emissions of N_2O from Scottish agricultural soils, as a function of fertilizer N. Nutr. Cycl. Agroecosyst. 52, 123–130.
Stehfest, E., Bouwman, A.L., 2006. N_2O and NO emission from agricultural fields and soils under natural vegetation: summarizing available measurement data and modeling of global annual emissions. Nutr. Cycl. Agroecosyst. 74, 207–228.
Sylvester-Bradley, R., Thorman, R.E., Kindred, D.R., Wynn, S.C., Smith, K.E., Rees, R.M., Topp, C.F.E., Pappa, V.A., Mortimer, N.D., Misselbrook, T.H., Gilhespy, S., Cardenas, L.M., Chauhan, M., Bennett, G., Malkin, S., Munro, D.G., 2015. Minimising Nitrous Oxide Intensities of Arable Crop Products. AHDB Cereals & Oils Project Report No. 548.
Syp, A., Faber, A., Kozak, M., 2016. Assessment of N_2O emissions from rapeseed cutlivation in Poland by various approaches. Int. Agrophys. 30, 501–507.
Teepe, R., Vor, A., Beese, F., Ludwig, A., 2004. Emissions of N_2O from soils during cycles of freezing and thawing and the effects of soil water texture, and duration of freezing. Eur. J. Soil Sci. 55, 357–365.
Van Groenigen, J.W., Kasper, G.J., Velthof, G.L., van den Pol-van Dasselaar, A., Kuikman, P.J., 2004. Nitrous oxide emissions from silage maize fields under different mineral nitrogen fertilizer and slurry applications. Plant Soil 263, 101–111.
Van Groenigen, J.W., Velthof, G.L., Oenema, O., van Groenigen, K.J., van Kessel, C., 2010. Towards an agronomic assessment of N_2O emissions. A case study for arable crops. Eur. J. Soil Sci. 61, 903–913.
WMO, 2016. The state of greenhouse gases in the atmosphere based on global observations through 2015. Greenh. Gas Bull. 12, 1–8.
Wagner-Riddle, C., Furon, A., McLaughlin, N.L., Lee, I., Barbeau, J., Jayasundara, S., Parkin, G., von Bertoldi, P., Warland, J., 2007. Intensive measurement of nitrous oxide emissions from a corn-soybean-wheat rotation under two contrasting management systems over 5 years. Global Chance Biol. 13, 1722–1736.
Walter, K., Don, A., Fuß, R., Kern, J., Drewer, J., Flessa, H., 2015. Direct nitrous oxide emissions from oilseed rape cropping – a meta-analysis. Glob. Change Biol. Bioenergy 7, 1260–1271.
Wood, S.N., 2011. Fast stable restricted maximum likelihood and marginal likelihood estimation of semiparametric generalized linear models. J. R. Stat. Soc. B 73, 3–36.
Wrage, N., Velthof, G.L., van Beusichem, M.L., Oenema, O., 2001. Role of nitrifier denitrification in the production of nitrous oxide. Soil Biol. Biochem. 33, 1723–1732.
Xu, X.K., Duan, C.T., Wu, H.H., Li, T.S., Cheng, W.G., 2016. Effect of intensity and duration of freezing on soil microbial biomass extractable C and N pools, and N_2O and CO_2 emissions from a forest soil in cold temperate region. Sci. China Earth Sci. 59, 156–169.
Zebarth, B.J., Rochette, P., Burton, D.L., 2008. N_2O emissions from spring barley production as influenced by fertilizer nitrogen rate. Can. J. Soil Sci. 88, 197–205.

5 Nitrous oxide emission and yield of winter oilseed rape fertilized with digestates and a nitrification inhibitor

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5.1 Abstract

Winter oilseed rape (OSR) is the major oil crop cultivated in Europe and the most important feedstock for biodiesel. Up to 90 % of the GHG emissions from biodiesel production can occur during OSR cultivation. Therefore, mitigation strategies for GHG emissions in the field are required and need to focus on direct nitrous oxide (N_2O) emission as one of the largest contributor. Substitution of synthetic N-fertilizers by digestates is currently under discussion due to the avoidance of GHG emissions during the production process of synthetic fertilizer. Since information on the effect of NIs on N_2O emissions from digestates is scare, the aim of this study was to evaluate their effect on N_2O emissions, mineral N dynamics, oil yield, and yield-related emissions in OSR production fertilized with digestate. The study was conducted at five different German sites over three years resulting in 15 full annual data sets. Over all sites and years, annual N_2O emission ranged between 0.2 and 3.5 kg N_2O-N ha^{-1} a^{-1}. A significant reduction of the N_2O emission after digestate application with NI was detected for three annual data sets. The mean reduction potential overall dataset was 36 %. During the fertilization period, N_2O emission varied between 0.01 and 1.8 kg N_2O-N ha^{-1} $period^{-1}$. In this period, NI reduced mean N_2O emission between 69 and 97 %.
Our results demonstrate that NI can be effective measure for reducing N_2O emissions from digestate application, but their effectiveness depends on soil and weather conditions and, ultimately, a sites potential for N_2O emission.

5.2 Introduction

During the last decade, demand of biofuel in Europe has been growing with rapeseed oil as the most important feedstock (Hamelinck et al. 2012; Carré and Pouzet 2014; Aldhaidhawi et al. 2017). As a result, oilseed rape (*Brassica napus* L., OSR) production in the European Union increased between 2000 and 2014 by 38 % (FAOSTAT 2017). However, there has long been a controversial discussion as to whether the climate impact of rapeseed cultivation is really positive. 75 to 90 % of the total GHG emissions during the production of biodiesel are emitted as nitrous oxide (N_2O) during the cultivation of feedstock in the field (66.7–119.5 g CO_2 MJ_{fuel}^{-1}) (Hoefnagels et al. 2010). Nitrous oxide contributes to the greenhouse effect (i.e., 100-year Global Warming Potential of 298; Mhyre et al. 2013) and to stratospheric ozone depletion (Crutzen 1981). 59 % of the entire anthropogenic N_2O emission is emitted from agricultural soils (IPCC 2006). Nitrification and biological denitrification are main sources for N_2O production in soils (Bremner 1997). Apart from these two processes, the contribution of further microbial and chemical N transformations, such as nitrifier-denitrification or chemo-denitrification, to the total N_2O release from soils is currently discussed (i.e. Shaw et al. 2006; Butterbach-Bahl et al. 2013). Since all processes of N_2O production in soils rely on mineral N, N-fertilization was frequently shown to enhance N_2O

emissions from agricultural soils (Stehfest & Bouwman 2006; Jungkunst et al. 2006; Kaiser and Ruser 2000).

To close the nutrient cycle within renewable energy production, digestate from biogas plants suggests itself as a substitute for mineral N-fertilizer. The number of biogas plants in Germany increased from 139 in 1992 to 9494 in 2018 (Statista 2018). Hereby digestate as valuable fertilizer gained importance. Anaerobic digestion changes the chemical composition of the biogas substrate, resulting in higher NH_4^+ contents, higher pH values and lower carbon contents (Möller and Müller 2012; Wolf et al. 2014). Fertilization with digestate is resulting in small-scale anaerobic zones due to the biological consumption of oxygen (O_2) (Zhu et al. 2015) and moisture retention by the organic matter of the digestate (Petersen et al. 2003) promoting N_2O hotspots through denitrification.

Nitrification inhibitors (NIs) inhibit the enzyme ammonia monooxygenase (AOM) which catalysis the first step of nitrification process carried out by microorganisms ($NH_4^+ \rightarrow NH_2OH$), thus stabilizing NH_4^+ stock. Many positive effects of NI application with organic fertilizer such as lower NO_3^- leaching, increased N-use efficiency and enhanced yields have been reported (Sutton et al. 1986; Di and Cameron 2007; Fangueiro et al. 2009). A meta-analysis from Abalos et al. (2014) showed an increase in productivity of different crops due to the use of NIs (DMPP; DCD; NBPT) where their effectiveness depended on environmental and management factors.

Since nitrification is one of the main sources of N_2O formation and since it provides NO_3^- as the substrate for N_2O release from denitrification, NIs are also a promising option for N_2O mitigation. With a mean of approximately 35 % N_2O reduction over all NIs (Akiyama et al. 2010; Ruser and Schulz 2015), fertilizer-derived N_2O emissions can be reduced up to 92 % (Menendez et al. 2012). However, the mitigation potential of NIs depends widely on site-specific conditions such as weather, soil properties and management practices (Volpi et al. 2017). Although numerous experiments studied the effect of NIs on N_2O emissions, only few studies investigated this effect for digestates. Wolf et al. (2014) reported a reduction of the N_2O emission between 37 and 62 % in the first weeks following application. Despite this high short-term mitigation potential, the effect was not significant on annual basis. In a laboratory incubation experiment Serverin et al. (2016) reported 70 % lower N_2O emissions from the treatment with 3,4-dimethyl pyrazole phosphate (DMPP) compared to untreated digestate.

Due to the sparse information on the effect of NIs on trace gas fluxes from OSR after digestate fertilization, the aim of the present work was to determine effects of NI application (1) on N_2O emissions and mineral N dynamics, (2) on grain and oil yield, and (3) on yield related emissions after digestate application in OSR production.

5.3 Material and Methods

5.3.1 Study sites, experimental design and management

This study was part of field trials at five experimental sites located in areas representative for OSR production in Germany (Table 1; Ruser et al. 2017). Since the main OSR cultivation area in Germany is located in the northern part, three sites where established in that area (Hohenschulen, Dedelow and Berge). One site was located in Middle Germany (Merbitz) and one in South Germany (Ihinger Hof). Soil and environmental characteristics of the study sites are shown in Table 5.1.

A randomized split-plot design with four replicate blocks was established at each site in 2012. Crop rotation with winter oilseed rape (var. 'Visby') – winter wheat (*Triticum aestivum* L., var. 'Julius') – winter barley (*Hordeum vulgare* L., var. 'Tenor' in Berge, var. 'Meridian' in Hohenschulen and var. 'Souleyka' at all other sites) was used as main plot factor. The main plots were managed according to best management practice at each site. Each main plot was subdivided into two subplots, one treated with and one without nitrification inhibitor. Plot size varied slightly across study sites due to different farming machinery; the minimum size was 3 x 9 m (27 m^2).

OSR was sown between end of August and early September at all sites and in all years (40 to 45 grains m^2, inter-row width 0.36 m). Fertilization was split into two doses as common for OSR production in Germany. The first dose was applied at the beginning of the growing season and the second approximately four weeks later. For each N-fertilization 90 kg NH_4 - N ha^{-1} was applied as digestate, resulting in a total of 180 kg NH_4-N ha^{-1} a^{-1}. Characteristics of the substrates are shown in Table 5.3. In 2014, the whole N amount was applied with only one single application at the study site Dedelow.

Piadin® (SKW, Piesteritz, Germany), which is a pyrazole derivative (PD), was used as nitrification inhibitor (5 l ha^{-1}) mixed with the digestates at all study sites. Piadin® contains two active compounds; the combination of 1H-1,2,4-triazole and 3-methylpyrazole has proved as an effective NI (Aulakh and Kuldip-Singh Doran 2001; Barneze et al. 2014; Wu et al. 2017). Trailing-hose application technique was used at all sites to apply the fertilizer between the plant rows. The only exception was Berge where an injection technique was used for the first 1st fertilizer dose in 2013. To avoid sulfur deficiency, 90 kg S ha^{-1} were applied to OSR as kieserite ($MgSO_4$) every spring. After harvest, soil was plowed at every site. The experimental design with three years of replicated N_2O emission measurements and crop yield determination at five sites enabled for a comprehensive analysis and comparison of NI effects at different sites and in different years.

5.3.2 Gas flux measurement

Between 2013 and 2015, N_2O flux rates were measured using the closed chamber method (Mosier & Hutchinson 1981) and calculated as described in detail by Ruser et al. (2017). Briefly, base frames (71 x 27 cm) were installed between the plant rows. Fluxes were measured at least once a week completed through additional event-oriented measurements (after fertilization, tillage, heavy rain, and during frost-thaw cycling). During gas sampling, dark closed chambers were placed airtight on the base frames. Four gas samples were taken periodically every 15-20 minutes out of the chambers' atmosphere and transferred into evacuated glass vials. N_2O and CO_2 concentration in the gas samples was analysed gas chromatographically with an electron capture detector (^{63}Ni-ECD). Fluxes were estimated by a procedure combining non-linear and robust linear flux models using the R (R Core Team 2017) package gasfluxes (Fuss 2016). Flux estimates were then submitted to rigorous quality checks by cross-checking with CO_2 accumulation, and missing or discarded fluxes were filled by multiple imputation (Honaker et al. 2011, Ruser et al. 2017).
Cumulative annual N_2O emissions were calculated for the periods between 1st January and 31st December. This definition for this period was chosen since it covered all soil management and N-fertilization measures of the OSR as well as the time when increased soil mineral N contents were expected. Furthermore, cumulative N_2O emission during the fertilization period was calculated. This period started with the date of the first digestate application and ended four weeks after the second N application (Table 5.2).
For the calculation of the cumulative N_2O emission, we linearly interpolated between two sampling dates. Oil yield-related N_2O emissions were calculated by relating the annual N_2O emissions to the amount of oil yield which was the product of OSR seed yield and oil concentration in the seeds.

Tab. 5.1: Main soil chemical and physical characteristics of the study sites.

Study site		Berge	Dedelow	Ihinger Hof	Hohenschulen	Merbitz
Coordinates		N 52° 61‘ 67‘‘ E 12° 78‘ 33‘‘	N 53° 36‘ 57‘‘ E 13° 82‘ 71‘‘	N 48° 73‘ 76‘‘ E 8° 92‘ 36‘‘	N 54° 31‘ 34‘‘ E 9° 99‘ 34‘‘	N 51° 61‘ 62‘‘ E 11° 91‘ 12‘‘
MAP 2013/14/15	[mm a^{-1}]	503 615/482/570	485 446/561/414	688 923/763/544	732 462/409/562	520 700/456/429
MAT 2013/14/15	[°C]	8.7 9.4/13.0/10.6	8.4 8.7/9.9/9.7	8.3 8.6/10.4/10.1	8.9 8.1/9.6/8.8	9.0 9.1/10.7/10.4
Clay	[%]	5.7	10.0	3.2	10.5	15.8
Silt	[%]	19.9	30.9	78.2	29.4	67.8
Sand	[%]	74.4	59.1	18.6	60.1	16.4
pH[§]	0.01 *M*	6.5	7.4	6.8	5.9	6.6
C_{org}[§]	[%]	1.15	0.75	1.68	1.87	1.18
N_t[§]	[%]	0.09	0.10	0.20	0.12	0.11
Soil type[$]		Luvisol	Luvisol	Haplic Luvisol	Haplic Luvisol/ Anthrosol	Haplic Chernosem

MAP: Long-term mean annual precipitation and annual precipitation in the single experimental years;
MAT: Long-term mean annual air temperature (2 m) and annual mean air temperature in the single experimental years, [§] measured in the top soil (0-30 cm). [$] IUSS Working Group WRB (2015).

Tab. 5.2: Calculation periods for statistical analysis.

Site	Year	1st Fertilization	2nd Fertilization		days
		start		end	
Berge	2013	04.04.	02.05.	30.05.	56
	2014	19.02.	09.04.	07.05.	77
	2015	17.02.	24.03.	21.04.	63
Dedelow	2013	05.03.	17.04.	15.05.	71
	2014	11.03.	11.03.	08.04.	28
	2015	03.03.	24.03.	21.04.	49
Ihinger Hof	2013	13.03.	15.04.	13.05.	61
	2014	25.02.	24.03.	21.04.	55
	2015	09.03.	13.04.	11.05.	63
Hohenschulen	2013	04.04.	23.04.	21.05.	47
	2014	12.03.	01.04.	29.04.	48
	2015	17.03.	08.04.	06.05.	50
Merbitz	2013	09.04.	02.05.	30.05.	51
	2014	10.03.	08.04.	06.05.	57
	2015	10.03.	08.04.	06.05.	57

5.3.3 Environmental, soil, and plant analyzes

Climate stations were installed at each study site directly next to the experimental plots. We detected air temperature in 2 and 0.05 m height and daily precipitation. Data loggers (LogTag, TRIX-8, CIK solutions, Karlsruhe, Germany) were used to determine soil temperature in 0.05, 0.1 and 0.2 m soil depth. Soil samples were taken simultaneously to each gas sampling from 0 to 0.3 m depth and pooled over the four replicate plots. For further analysis, the samples were sieved (< 5 mm) and stored frozen. To determine the mineral N content, 80 g of fresh soil were extracted with 200 ml of a 1.25 10^{-2} *M* $CaCl_2$ solution. Photometric flow-injection analyzers were used to measure the concentrations of NO_3^- and NH_4^+ in the extracts. Comparability of the different analyzers used for that purpose was established in an inter-laboratory comparison test. Soil moisture was determined gravimetrically by drying soil at 105°C for 24 h. Bulk density of the topsoil was determined using stainless steel cylinders (100 ml) before and after each soil management event. Oil content of the OSR seeds was measured using near-infrared spectroscopy (NIRSystem 5000, Foss, Hamburg, Germany).
In 2017, the German Fertilizer Ordinance, which regulates the maximum amount of N-fertilizer, was revised. In contrast to the preceding ordinance (DüV, 1996) the maximum amount of N applied with digestate in arable land was reduced from 170 kg N ha^{-1} based on NH_4-N to 170 kg N ha^{-1} based on total N (DüV, 2017).

Tab. 5.3: Main characteristics of the digestates used at the different study sites.

Site		Berge			Dedelow			Ihinger Hof			Hohenschulen			Merbitz		
		2013	2014	2015	2013	2014	2015	2013	2014	2015	2013	2014	2015	2013	2014	2015
Substrate		cattel slurry/ maize/grass silage			cattle slurry/ maize silage			pig slurry/ maize/grass silage			cattle slurry/ maize silage			cattle slurry/ maize silage		
pH		8.1	7.9	7.6	8.1	7.6	8.7	8.0	7.9	7.9	8.0	8.1	8.1	7.7	7.9	7.7
DM	[%]	8.4	9.2	10.5	6.0	6.1	7.8	8.9	10.1	8.7	6.8	7.1	8.1	5.2	5.1	5.0
OM	[%]	6.3	6.1	6.2	4.5	4.2	~	6.5	7.1	5.9	~	~	~	3.6	3.5	3.3
C_{org}	[%]	3.7	3.5	3.6	2.3	2.4	3.0	3.8	4.1	3.4	~	~	~	2.1	2.0	1.9
C_t	[%]	~	~	~	2.6	2.4	3.0	3.3	4.0	3.4	~	~	~	~	~	~
N_t	[%]	0.6	0.5	0.5	0.3	0.3	0.5	0.5	0.6	0.6	0.4	0.5	0.5	0.3	0.3	0.3
NH_4-N	[%]	0.4	0.2	0.2	0.1	0.2	0.2	0.3	0.3	0.4	0.3	0.3	0.3	0.2	0.2	0.2
NH_4-N applied	[kg ha^{-1}]	180	180	180	129	191	184	171	190	197	174	164	197	180	180	180

DM = dry matter; OM = organic matter; ~ = not determined/ not calculated.

5.3.4 Statistical methods

Normal distribution and variance homogeneity of residuals were checked graphically. To reach normal distribution and homogeneous variances of residuals for N_2O and CO_2 emissions, a logarithmic transformation of the data was carried out prior to analysis.
A mixed model approach using SAS PROC MIXED for the comparison of cumulative N_2O emissions, oil yield and oil yield-related N_2O emissions was used. Last square means were calculated and compared with an LSD-test at $\alpha = 0.05$.
For each site, a multiple regression analysis for nitrous oxide fluxes was performed. To model block effects and their inclusion in the multiple regression approach, dummy variables were created. They were included by default in the model. A stepwise forward selection was used in the multiple regression approach with $\alpha = 0.05$ on the following explaining variables: air temperature (2 m), NO_3, water-filled pore space (WFPS) and CO_2. From this approach, the correlation was determined as the square root of the partial R^2 value. As only significant explaining variables remain in the model, all explaining variables in the final model show significant effects. Analyses were performed in SAS 9.4 (SAS Institute, Cary NC). Plots and graphics were created with SigmaPlot 11.0 (Systat Software GmbH, Erkrath).

5.4 Results and Discussion

5.4.1 Meteorological conditions

The annual precipitation varied from 409 mm (Hohenschulen, 2014) to 923 mm (Ihinger Hof, 2013) (Table 5.1). At each study site, the long-term mean annual precipitation lay within the range of the annual precipitation in the three single experimental years indicating, that our measurements were conducted in a period with site representative precipitation characteristics. At Ihinger Hof and Merbitz the annual precipitation in 2013 was 34 and 35 % higher when compared to the long-term mean. Also in 2013, Berge had 22 % higher precipitation than the long-term mean. Particularly spring and summer 2015 was very dry at almost every site.
Mean air temperature ranged between 8.1°C (Hohenschulen, 2013) and 13.0°C (Berge, 2014). With the exception of the study site Hohenschulen, annual mean temperature was predominately higher than the long-term mean at all remaining study sites and in all three experimental years.

5.4.2 N_2O fluxes

N_2O fluxes showed a high spatial and temporal variability (Figures 5.1 and 5.2). Increased fluxes were often measured after digestate application in conjunction with precipitation events. In May 2013, the highest flux rate during the entire experimental period was

determined with 458 µg N_2O-N m^{-2} h^{-1} after second fertilization following a heavy rain shower (42 mm d^{-1}) at the study site Merbitz (-NI) (Figure 5.1). High N_2O flux rates after N-fertilization were also measured 2013 in Berge, 2015 in Dedelow, and 2014 and 2015 in Hohenschulen.

High spatial and temporal variability of N_2O fluxes has frequently been reported from arable soils with increased flux rates after N-fertilization and rainfall and mainly explained with enhanced denitrification (Ambus and Christensen 1994; Flessa et al. 1995; Kaiser et al. 1996). After fertilization, ammonium in soils is rapidly oxidized to nitrate (Singh and Verma 2007) thus increasing substrate availability for denitrification. Further, denitrification is enhanced by the formation of anaerobic conditions as result of lower gas diffusivity in soil water when compared to soil air (Heincke & Kaupenjohann, 1999) and therefore of a reduced O_2 diffusion into soil combined with high microbial O_2 demand during C turnover (Flessa et al. 1995; Corre et al. 1996; MacKenzie et al. 1997).

Winter fluxes were generally low at all sites and in all years. Due to the absence of severe frost/thaw cycling over the entire experimental time, there were no considerable frost/thaw induced N_2O pulses at our study sites (Ruser et al. 2017).

For every experimental site, N_2O fluxes were significant correlated ($p < 0.0001$, Table 5.4) with CO_2 fluxes. The measured CO_2 fluxes are an indicator for heterotrophic microbial activity during decomposition of soil organic matter, where the microbial activity also includes the activity of denitrifiers in soils. Except for the sandy site Berge, we also found a positive correlation between the N_2O fluxes and soil moisture. Additionally, N_2O fluxes in Dedelow, Hohenschulen and Merbitz were also positive correlated with soil nitrate contents (Table 5.4). Enhanced N_2O flux rates with increasing soil moisture and in part with nitrate contents suggest denitrification as a major N_2O source over all study sites.

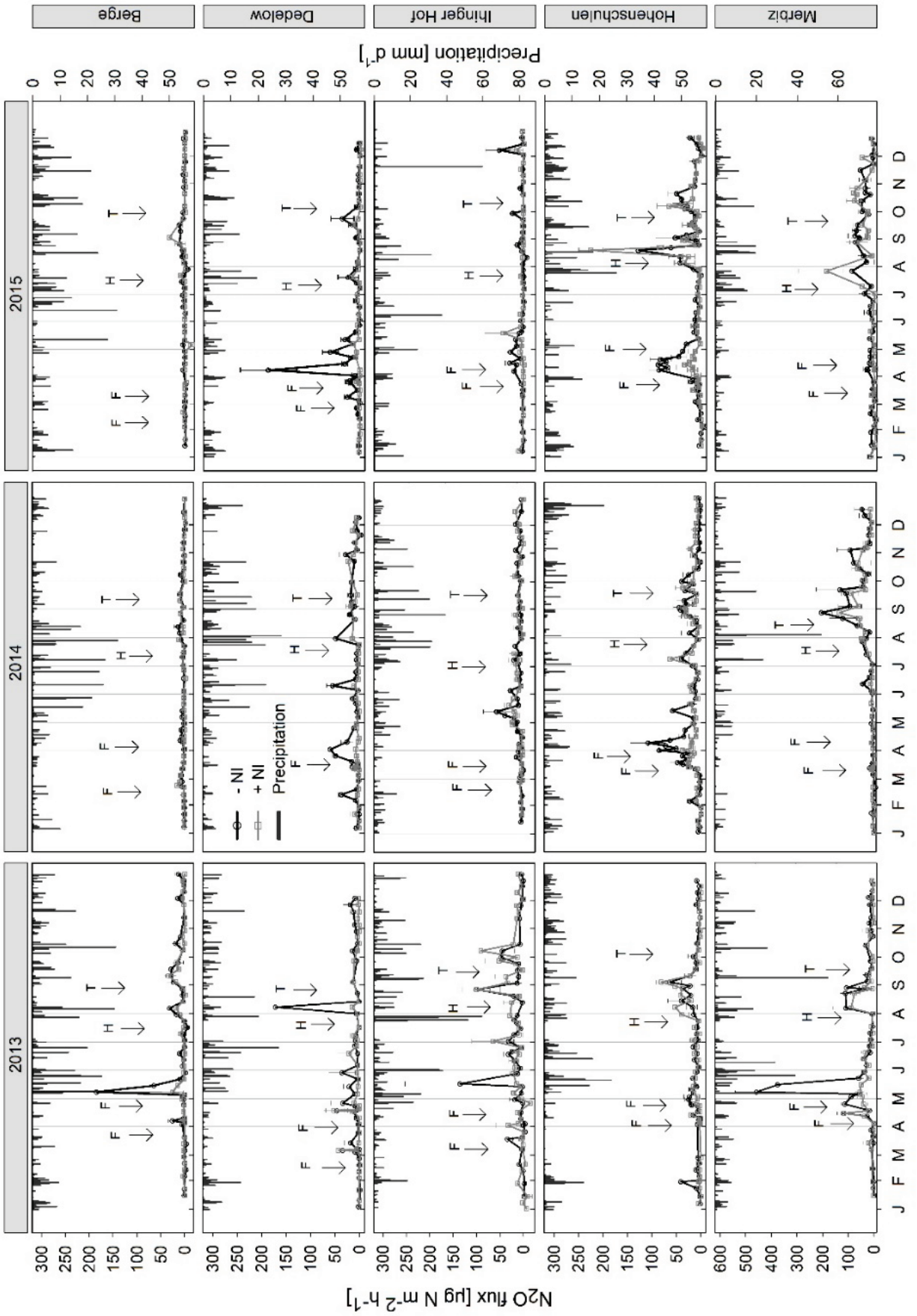

Fig. 5.1: Temporal pattern of the mean N_2O flux rate (n = 4) of the treatments +NI and -NI and daily rainfall as affected by study site and experimental year. Note: different y-axis scaling.

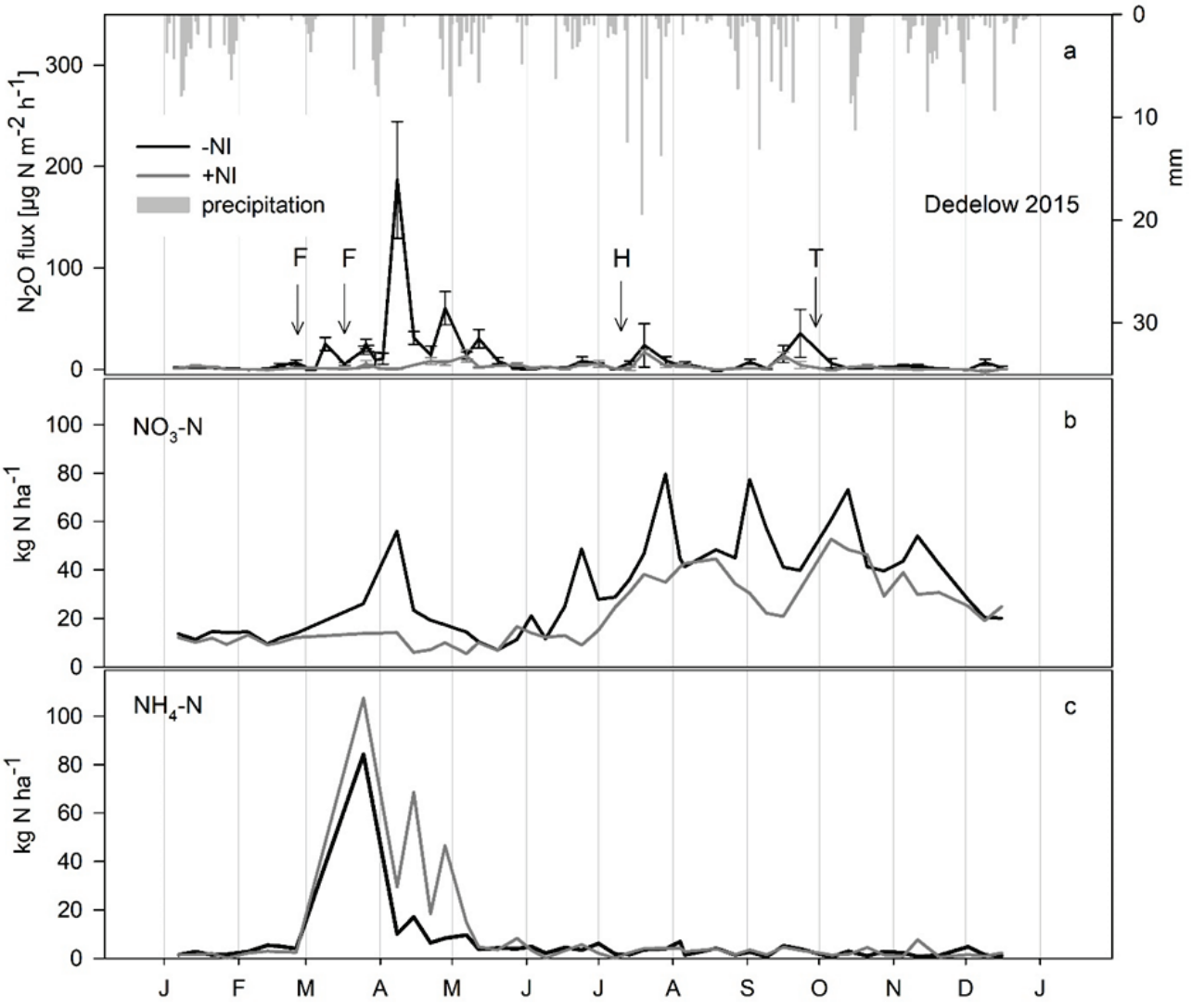

Fig. 5.2: Temporal pattern of the mean N_2O flux rate (n = 4) of the treatments +NI and -NI (a) and daily rainfall at Dedelow 2015. NO_3-N (b) and NH_4-N (c) content of the topsoil (0-30 cm) affected by NI application.

Tab. 5.4: Regression analysis (PROC REG stepwise forward regression) for N_2O fluxes by site. CO_2, NO_3-N, WFPS and temperature (2 m height) were parameters included.

Study site	estimate	R^2					F-Value	p-value
		CO_2	NO_3	WFPS	T	∑		
Berge	0.00077	0.06				*0.06*	32.1	<.0001
Dedelow	0.00144	0.14					104.0	<.0001
	0.00230			0.02		*0.17*	15.4	<.0001
	0.00126		0.01				11.1	0.0009
Ihinger Hof	0.00102	0.21					148.2	<.0001
	0.00146			0.01		*0.23*	8.0	0.0049
	0.00397				0.01		4.6	0.0032
Hohenschulen	0.00200	0.29					238.2	<.0001
	0.00386		0.06			*0.37*	31.2	<.0001
	0.00289			0.02			13.2	0.0003
Merbitz	0.00163	0.17					209.1	<.0001
	0.01646				0.04	*0.26*	48.8	<.0001
	0.00252		0.03				40.3	<.0001
	0.00793			0.02			26.4	<.0001

5.4.3 NI effects on N_2O fluxes, cumulative N_2O emission and mineral N

Figure 5.2 shows exemplarily the N_2O fluxes from study site Dedelow in 2015. It illustrates the inhibitory effect of NI application. Following digestate applications at this site in 2015, N_2O fluxes were significantly reduced when the NI was added to the digestate.

Cumulative N_2O emissions during the fertilization period varied between 0.01 kg N_2O-N ha^{-1} $period^{-1}$ and 1.8 kg N_2O-N ha^{-1} $period^{-1}$ (Table 5.5). In 12 of the 15 data sets, mean N_2O emission was lower when NIs were applied along with the digestates, with this effect being statistically significant in six data sets. Generally, N_2O reduction during this period in the statistically significant data stets was very high; it varied between 69 % (Ihinger Hof in 2015) and 97 % (Dedelow, 2014).

As mentioned NIs inhibit ammonia monooxygenase (AMO) as the first step during nitrification and therefore directly decrease the release of N_2O from nitrification. Due to lower nitrate concentration as substrate for denitrifiers, NIs also reduce N_2O emissions from denitrification (Ruser and Schulz 2015). Additionally, the N_2/N_2O-ratio increases with decreasing NO_3^- concentration as result of a competitive effect of NO_3^- and N_2O as terminal electron acceptor during denitrification (Blackmer and Bremner 1978; Ruser et al. 2006). This would further decrease N_2O release from denitrification.

Following digestate application the NH_4-N/NO_3-N ratio in the treatment +NI at the study sites Dedelow, Ihinger Hof and Hohenschulen was higher than in the treatment without NI, this was also measured at the site Berge in the third year (Figures 5.1 and 5.3). When compared to the -NI treatments, higher NH_4-N/NO_3-N ratios in the +NI treatments indicate the inhibitory effect of NIs. This effect lasted between one and four weeks after digestate application. For the study site Merbitz (loamy silty texture) this effect could not be observed. This was explained with the low extraction efficiency of 0.0125 *M* $CaCl_2$ solution for NH_4^+ on loamy

and silty soils (Kuderna et al. 1993) disabling the use of this ratio as indicator for the inhibition of nitrification.

As result of the inhibitory effect, nitrate availability following fertilization was lower when compared to treatments without NI application (Figure 5.2). Even the mean annual nitrate contents of the +NI treatments were lower than the ones in the -NI treatments at every site and in every experimental year (except for Hohenschulen 2015; Table 5.6).

Total annual N_2O emission varied between 0.2 kg N_2O-N ha^{-1} a^{-1} and 3.5 kg N_2O-N ha^{-1} a^{-1} (Table 5.5). The same range of annual N_2O emissions was reported for OSR fertilized with mineral N fertilizer at the same study sites and in the same experimental period (Ruser et al. 2017). They mainly explained the high inter-annual variability of the N_2O emission at our study site with varying weather conditions between the experimental years with rainfall as the main driver for increased N_2O fluxes after N-fertilization measures.

The mean annual N_2O emission decreased in the following order: Merbitz (2.8 kg) > Hohenschulen (1.5 kg) > Dedelow (1.1 kg) > Ihinger Hof (1.0 kg) > Berge (0.5 kg N_2O-N ha^{-1} a^{-1}) (Table 5.5). Ruser et al. (2017) also reported almost the same order for annual emissions from mineral N-fertilization increasing from sandy to loamy textures of the top soils and with increasing C_{org} contents at our five study sites. Sandy soil texture and low C_{org} contents result in a low water holding capacity and thus in good soil aeration limiting denitrification (Bouwman et al. 2002). Pelster et al. (2012), Stehfest and Bouwman (2006) and Leip et al. (2011) have reported similar results for N_2O emissions from soils with varying soil texture.

When compared to the treatment without NI, mean annual N_2O emission in the treatment with NI was lower in 10 of the 15 full annual data sets with three of these 10 sets being statistically significant ($p < 0.05$, Table 5.5).

The mean annual emission over all data was 1.0 kg N_2O-N ha^{-1} a^{-1} in the treatment -NI and significantly lower when digestate was applied together with NI (0.6 kg N_2O-N ha^{-1} a^{-1}). Despite of the generally low fluxes, the use of NI reduced the mean annual N_2O emissions over the whole data set by 36 %. This reduction potential was in good agreement with studies which summarized the results of experiments on the effect of NIs on N_2O emissions from agricultural soils (Akiyama et al. 2010; Ruser and Schulz 2015).

So far, no other agricultural measure has so consistently proofed such a high reduction potential for N_2O emissions in the field than the application of NIs. The discrepancy between the reduction potential calculated for the fertilization period (54 %) and for the data on annual basis (36 %) as well as the lower amount of statistically significant data sets on annual base clearly emphasize the need of a uniform observation period for a reliable evaluation of measures to reduce greenhouse gas emissions from agricultural soils. Several authors suggested full annual measurements for that purpose (Bouwman 1996; Ruser et al. 2001).

Tab. 5.5: N_2O emissions annual and during fertilization period, oil yield and oil yield-related N_2O emissions effected by site and year and nitrification inhibitor (+NI; -NI). Statistically significant differences between +NI and –NI treatment within single year and within single study site was indicated by small letters (LSD, p = 0.05).

Study site	Exp. year	N_2O emission				Oil yield		Oil yield-related N_2O emissions	
		annual		fertilization					
		[kg N_2O-N ha^{-1} a^{-1}]		[kg N_2O-N ha^{-1}]		[Mg ha^{-1} a^{-1}]		[kg N_2O-N Mg^{-1} oil ha^{-1}]	
		- NI	+ NI	- NI	+ NI	- NI	+ NI	- NI	+ NI
Berge	2013	1.1 [a]	0.4 [a]	0.42 [a]	0.08 [a]	1.3 [a]	1.2 [a]	0.8 [a]	0.3 [a]
	2014	0.3 [a]	0.2 [a]	0.07 [a]	0.05 [a]	1.8 [a]	1.8 [a]	0.2 [a]	0.1 [a]
	2015	0.2 [a]	0.2 [a]	0.01 [a]	0.01 [a]	1.5 [a]	1.4 [a]	0.1 [a]	0.2 [a]
	2013-2015	*0.5*	*0.3*			*1.5*	*1.5*	*0.4*	*0.2*
Dedelow	2013	1.1 [a]	0.5 [a]	0.22 [a]	0.09 [a]	2.7 [a]	2.6 [a]	0.4 [a]	0.2 [a]
	2014	**1.2 [a]**	**0.6 [b]**	**0.15 [a]**	**0.03 [b]**	2.6 [a]	2.6 [a]	**0.5 [a]**	**0.2 [b]**
	2015	**1.1 [a]**	**0.3 [b]**	**0.44 [a]**	**0.01 [b]**	2.1 [a]	2.1 [a]	**0.5 [a]**	**0.1 [b]**
	2013-2015	*1.1*	*0.5*			*2.5*	*2.4*	*0.5*	*0.2*
Ihinger Hof	2013	1.4 [a]	1.7 [a]	0.06 [a]	0.10 [a]	1.9 [a]	1.8 [a]	0.8 [a]	1.0 [a]
	2014	0.9 [a]	1.2 [a]	0.06 [a]	0.07 [a]	1.8 [a]	1.6 [a]	0.5 [a]	0.7 [a]
	2015	0.5 [a]	0.4 [a]	**0.19 [a]**	**0.03 [b]**	1.9 [a]	1.8 [a]	0.3 [a]	0.2 [a]
	2013-2015	*1.0*	*1.1*			*1.8*	*1.8*	*0.5*	*0.6*
Hohenschulen	2013	1.0 [a]	1.0 [a]	0.12 [a]	0.09 [a]	2.1 [a]	2.1 [a]	0.5 [a]	0.5 [a]
	2014	1.5 [a]	1.2 [a]	**0.47 [a]**	**0.19 [b]**	2.5 [a]	2.5 [a]	0.6 [a]	0.5 [a]
	2015	1.9 [a]	1.3 [a]	**0.49 [a]**	**0.15 [b]**	2.1 [a]	2.0 [a]	0.7 [a]	0.6 [a]
	2013-2015	*1.5*	*1.1*			*2.2*	*2.2*	*0.6*	*0.5*
Merbitz	2013	**3.5 [a]**	**1.8 [b]**	**1.82 [a]**	**0.51 [b]**	1.9 [a]	1.8 [a]	**1.9 [a]**	**1.0 [b]**
	2014	2.6 [a]	1.9 [a]	0.04 [a]	0.03 [a]	2.1 [a]	2.1 [a]	1.2 [a]	0.9 [a]
	2015	2.2 [a]	2.2 [a]	0.16 [a]	0.03 [a]	1.7 [a]	1.6 [a]	1.4 [a]	1.3 [a]
	2013-2015	*2.8*	*2.0*			*1.9*	*1.8*	*1.5*	*1.1*

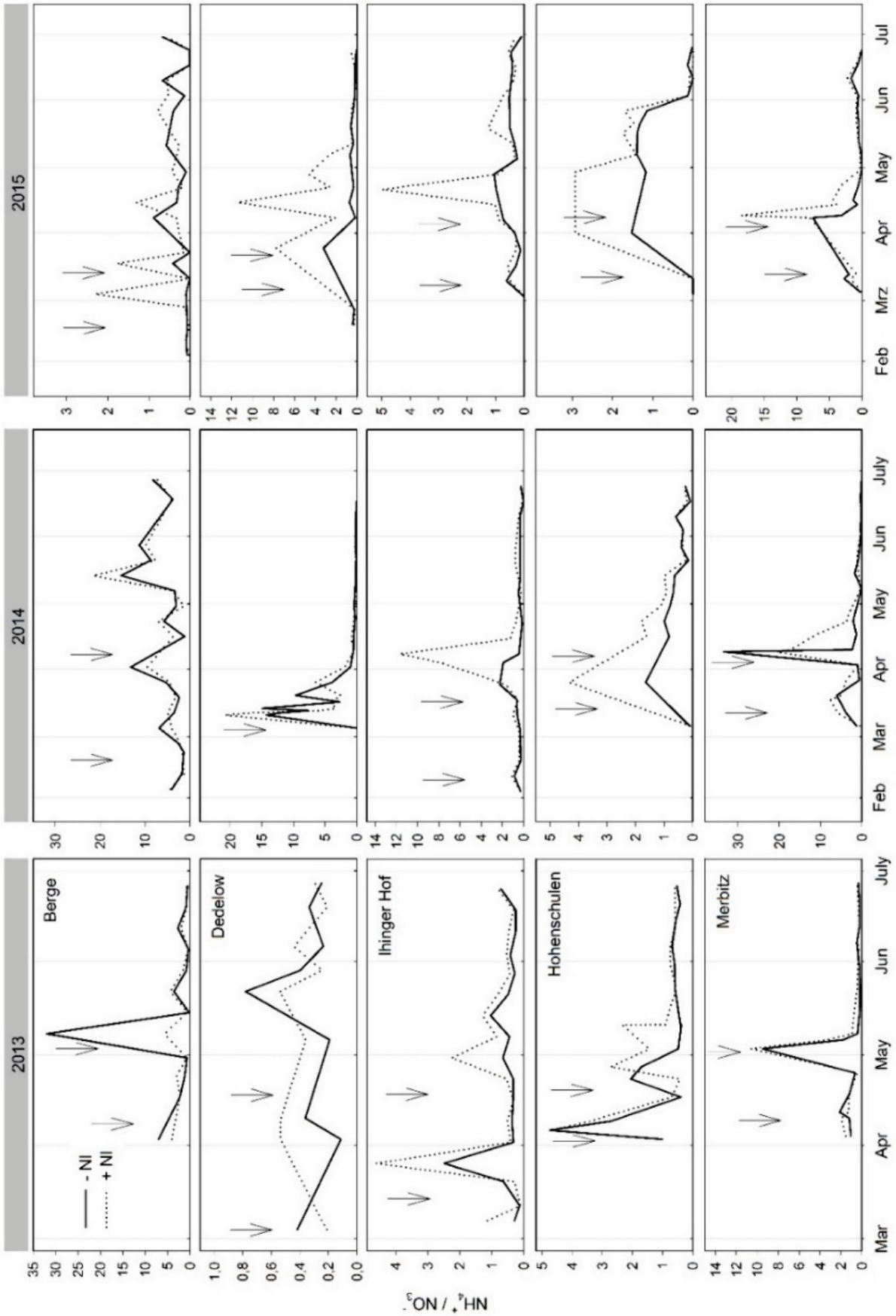

Fig. 5.3: Mineral NH_4^+ to NO_3^- ratio (0-30 cm depth) after digestate application as affected by site and experimental year.

Tab. 5.6: Mean annual NO_3^- - N (kg N ha^{-1}) content in topsoil (0-30 cm) as affected by nitrification inhibitor application, study site and experimental year.

Site	NO_3^- - N content in topsoil					
	2013		2014		2015	
	-NI	+NI	-NI	+NI	-NI	+NI
Berge	30.8	24.0	6.1	5.8	15.1	15.0
Dedelow	18.0	16.4	11.2	10.6	21.2	10.9
Ihinger Hof	11.6	10.9	15.1	12.3	17.7	15.1
Hohenschulen	20.2	18.4	12.9	10.1	22.9	24.0
Merbitz	7.9	6.7	5.5	5.2	8.5	6.6

5.4.4 NI effects on grain yield, oil yield and oil yield-related N_2O emission

The OSR grain yield was not affected by NI application (data not shown). On a three years average it ranged between 3.1 Mg ha^{-1} a^{-1} and 5.2 Mg ha^{-1} a^{-1}. The yields measured at our investigation sites were in the same range as the mean German OSR yields; 4.0, 4.5, and 3.9 Mg ha^{-1} a^{-1} in 2013, 2014, and 2015, respectively (German Federal Statistical Office, 2017). Mean OSR oil yield over the entire experiment was 2.0 Mg ha^{-1} a^{-1} (Table 5.5). It ranged between 1.2 and 2.7 Mg ha^{-1} a^{-1} and it was not affected by NI application (Table 5.5).
In a study on the effect of different preceding crops on OSR yield, Hegewald et al. (2016) reported only small increases in oil yield (0.04 Mg ha^{-1} a^{-1}) when N fertilization was increased from 120 to 180 kg N ha^{-1} a^{-1}. As mentioned by Rathke et al. (2006) N fertilization increases the crude protein content of rapeseeds at the expense of oil concentration. Since we fertilized 180 kg N ha^{-1} a^{-1} in our experiment, we considerably exceeded the N amount required for maximum oil yield and therefore an effect of NI on oil yield could not be expected.
Consistent with our study Wolf et al. (2014) also found no effect of NI on the yield of digestate fertilized maize. In other studies, higher yield was the result of NI application combined with an increased N use efficiency (Ladha et al. 2005; Alonso-Ayuso et al. 2016).
Oil yield-related emission varied between 0.1 and 1.9 kg N_2O-N Mg^{-1} oil ha^{-1} (Table 5.5). With few exceptions, the oil yield-related N_2O emissions in the +NI treatments were lower than in the -NI treatments. The differences between the N_2O emissions from these two treatments were statistically significant ($p < 0.05$) at Merbitz 2013, Dedelow 2014 and 2015. In Merbitz this significant effect could be explained with the significant lower N_2O emission in the -NI treatment whereas the oil yield was the same in both treatments. This was also the case for Dedelow 2014 and 2015 although NI at these sites and years did not significantly reduce N_2O emissions.
Significant lower yield-scaled N_2O emissions as result of NI applications were measured in maize and wheat (Gao and Bian 2017). In contrast to our experiment, the significant effects reported by Gao and Bian (2017) were the result of increased crop yields. Publications about yield-scaled N_2O emission from crops fertilized with digestate affected by NI are sparse. Studies with mineral fertilization showed a linear increase of yield-scaled N_2O emissions with increasing N fertilization rate (Lebender et al. 2014; Ruser et al. 2017).

5.5 Conclusion

Mitigation effect of NI after digestate application was not observed at every site and year. However, although not statistically significant in every set, the mean N_2O reduction of the total data sets was 36 % confirming a high reduction potential in years where environmental and management conditions allowed for NI effect. In years where the conditions were not favorable for N_2O reduction after NI application, NI application did not negatively influence OSR yield or oil yield related N_2O emission.
Comparing the NI effect on cumulative N_2O emission during the fertilization period (6 statistically significant data sets) with the effect on an annual base (3 statistically significant data sets), it gets obvious, that full annual data sets are needed for a reliable evaluation of the effect of measures on N_2O emissions from agricultural fields.
It has been shown, that surface application of NH_4^+ based fertilizers with NI can result in increased ammonia (NH_3) emissions (Kim et al. 2012). For a better environmental assessment of the effect of NIs, further investigation on trace gas fluxes from fields with NI application should therefore also consider NH_3 losses.

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5.6 References

Akiyama H, Yan X, Yagi K (2010) Evaluation of effectiveness of enhanced–efficiency fertilizers as mitigation options for N_2O and NO emissions from agricultural soils. Meta-analysis. Glob Change Biol 16:1837–1846

Aldhaidhawi M, Chiriac R, Badescu V (2017) Ignition delay, combustion and emission characteristics of Diesel engine fueled with rapeseed biodiesel – A literature review. Renew Sust Energ Rev 73:178–186

Ambus P and Christensen S (1994) Measurement of N_2O emission from a fertilized grassland: An analysis of spatial variability. J Geophys Res 99:16549–16555

Aulakh MS, Kuldip-Singh, Doran J (2001) Effects of 4–amino 1,2,4–triazole, dicyandiamide and encapsulated calcium carbide on nitrification inhibition in a subtropical soil under upland and flooded conditions. Biol Fertil Soils 33:258–263

Barneze AS, Minet EP, Cerri CC, Misselbrook T (2014) The effect of nitrification inhibitors on nitrous oxide emissions from cattle urine depositions to grassland under summer conditions in the UK. Chemosphere 119:122–129

Blackmer AM, Bremner JM (1978) Inhibitory effect of nitrate on reduction of N_2O to N_2 by soil microorganisms. Soil Biol Biochem 10:187–191

BMEL (2017) Novelle der Düngeverordnung: Düngen nach guter fachlicher Praxis. https://www.bmel.de/DE/Landwirtschaft/Foerderung-Agrarsozialpolitik/_Texte/Duengung-Cross-Compliance.html. Accessed 6 January 2018

BMJV (2017) Verordnung über die Anwendung von Düngemitteln, Bodenhilfsstoffen, Kultursubstraten und Pflanzenhilfsmitteln nach den Grundsätzen der guten fachlichen Praxis beim Düngen. https://www.gesetze-im-internet.de/d_v_2017/anlage_3.html. Accessed 6 January, 2018

Bouwman AF (1996) Direct emission of nitrous oxide from agricultural soils. Nutr Cycl Agroecosyst 46:53–70

Bouwman AF, Boumans LJM, Batjes NH (2002) Emissions of N_2O and NO from fertilized fields: summary of available measurement data. Glob Biogeochem Cycles 16:1058

Bremner J M (1997) Sources of nitrous oxide in soils. Nutr Cycl Agroecosyst 49:7–16

Burchill W, Li D, Lanigan GJ, Williams M, Humphreys J (2014) Interannual variation in nitrous oxide emissions from perennial ryegrass/white clover grassland used for dairy production. Glob Change Biol 20:3137–3146

Butterbach-Bahl K, Baggs EM, Dannenmann M, Kiese R, Zechmeister-Boltenstern S (2013) Nitrous oxide emissions from soils. How well do we understand the processes and their controls? Philos T Roy Soc B 368:20130122

Carré P and Pouzet A (2014) Rapeseed market, worldwide and in Europe. OCL 21, D102

Clayton H, McTaggart I, Parker J, Swan L, Smith KA (1997) Nitrous oxide emissions from fertilised grassland: A 2-year study of the effects of N fertiliser form and environmental conditions. Biol Fertil Soils 25:252–260

Corre MD, Van Kessel C, Pennock DJ (1996) Landscape and seasonal patterns of nitrous oxide emissions in a semiarid region. Soil Sci Soc Am J 60:1806–1815

Crutzen PJ (1981) Atmospheric chemical processes of the oxides of nitrogen including nitrous oxide. In: Delwiche, C.C. (Ed.), Denitrification, Nitrification and Atmospheric N_2O. Wiley, Chichester, pp. 17–44

Di HJ and Cameron KC (2007) Nitrate leaching losses and pasture yields as affected by different rates of animal urine nitrogen returns and application of a nitrification inhibitor–a lysimeter study. Nutr Cycl Agroecosyst 79:281–290

Dobbie KE, McTaggart IP, Smith KA (1999) Nitrous oxide emissions from intensive agricultural systems: variations between crops and seasons; key variables; and mean emission factors. J Geophys Res 104:26891–26900

FAOSTAT (2017) http://www.fao.org/faostat/en/#data. Accessed 18 August 2017

Fangueiro D, Fernandes A, Coutinho J, Moreira N, Trindade H (2009) Influence of two nitrification inhibitors (DCD and DMPP) on annual ryegrass yield and soil mineral N dynamics after incorporation with cattle slurry. Commun Soil Sci Plant Anal 40:3387–3398

Flessa H, Dörsch P, Beese F (1995) Seasonal variation of N_2O and CH_4 fluxes in differently managed arable soils in southern Germany. J Geophys Res 100:23115–23124

Gao W and Bian X (2017) Evaluation of the agronomic impacts on yield-scaled N_2O emission from wheat and maize fields in China. Sustainability 9:1201

Hamelinck C, De Loveinfosse I, Koper M (2012) Renewable energy progress and biofuels Sustainability. Ecofys, London, pp 450

Hegewald H, Koblenz B, Wensch-Dorendorf M, Christen O (2016) Impacts of high intensity crop rotation and N management on oilseed rape productivity in Germany. Crop Pasture Sci 67:439–449

Heincke M, Kaupenjohann M (1999) Effects of soil solution on the dynamics of N_2O emissions: a review. Nutr Cycl Agroecosys 55:133–157

Hoefnagels R, Smeets E, Faaij A (2010) Greenhouse gas footprints of different biofuel production systems. Renew Sustainable Energy Rev 14:1661–1694

Honaker J, King G, Blackwell M (2011) Amelia II: A program for missing data. J Stat 772 Softw 45:1–47

IPCC–Intergovernmental Panel on Climate Change (2006) Guidelines for national greenhouse gas inventories. Available at http://www.ipcc-nggip.iges.or.jp/public/2006gl. Geneva, Switzerland. Accessed 11 Mai 2017

Jungkunst HF, Freibauer A, Neufeldt H, Bareth G (2006) Nitrous oxide emissions from agricultural land use in Germany – a synthesis of available annual field data. J Plant Nutr Soil Sci 169:341–351

Kaiser EA and Ruser R (2000) Nitrous oxide emissions from arable soils in Germany – An evaluation of six long-term field experiments. J Plant Nutr Soil Sci 163:249–259

Kaiser EA, Eiland F, Germon JC, Gispert MA, Heinemeyer O, Henault C, Lind AM, Maag E, Saguer E, Van Cleemput O, Vermoesen A, Webster C (1996) What predicts nitrous oxide emissions and denitrification N–loss from European soils? J Plant Nutr Soil Sci 159:541–547

Kim DG, Saggar S, Roudier P (2012) The effect of nitrification inhibitors on soil ammonia emissions in nitrogen managed soils: A meta-analysis. Nutr Cycl Agroecosys 93:51–6

Kuderna M, Pötsch E, Blum WEH (1993) Zur Wahl des Extraktionsmittels bei der Nmin-Bestimmung. Bodenkultur 44:7–14

Ladha JK, Pathak H, Krupnik TJ, Six J, van Kessel C (2005) Efficiency of fertilizer nitrogen in cereal production: retrospects and prospects. Adv Agron 87:85–156

Lebender U, Senbayram M, Lammel J, Kuhlmann H (2014) Impact of mineral N fertilizer application rates on N_2O emissions from arable soils under winter wheat. Nutr Cycl Agroecosyst 100:111–120

Leip A, Busto M, Winiwarter W (2011) Developing spatially stratified N_2O emission factors for Europe. Environ Poll 159:3223–3232

MacKenzie AF, Fan MX, Cadrin F (1997) Nitrous oxide emission as affected by tillage, corn-soybean-alfalfa and nitrogen fertilization. Can J Soil Sci 77:145–152

Menendez S, Barrena I, Setien I, Gonzalez-Murua C, Estavillo JM (2012) Efficiency of nitrification inhibitor DMPP to reduce nitrous oxide emissions under different temperature and moisture conditions. Soil Biol Biochem 53:82–89

Mosier AR, Parton WJ, Hutchinson GL (1983) Modelling nitrous oxide evolution from cropped and native soils. In: Hallberg, R. (Ed.), Environmental Biogeochemistry. Ecological Bulletin, Stockholm, 229–241

Hutchinson GL and Mosier AR (1981) Improved Soil Cover Method for Field Measurement of Nitrous Oxide Fluxes. Soil Sci Soc Am J 45:311–316

Möller K and Müller T (2012) Effects of anaerobic digestion on digestate nutrient availability and crop growth. A review. Eng Life Sci 12:242–257

Pelster DE, Chatigny MH, Rochette P, Angers DA, Rieux C, Vanasse A (2012) Nitrous oxide emissions respond differently to mineral and organic nitrogen sources in contrasting soil types. J Environ Qual 41:427–435

Pfab H, Palmer I, Buegger F, Fiedler S, Müller T, Ruser R (2012) Influence of a nitrification inhibitor and of placed N-fertilization on N_2O fluxes from a vegetable cropped loamy soil. Agric Ecosyst Environ 150:91–101

Rathke GW, Behrens T, Diepenbrock W (2006) Integrated nitrogen management strategies to improve seed yield, oil content and nitrogen efficiency of winter oilseed rape (*Brassica napus* L.): a review. Agric Ecosyst Environ 117:80–108

Reeves S and Wang W (2015) Optimum sampling time and frequency for measuring N_2O emissions from a rain-fed cereal cropping system. Sci Total Environ 530-531:219–226

Ruser R and Schulz R (2015) The effect of nitrification inhibitors on the nitrous oxide (N_2O) release from agricultural soils–a review. J Plant Nutr Soil Sc 178:171–188

Ruser R, Flessa H, Schilling R, Beese F, Munch JC (2001) Effect of crop-specific field management and N fertilization on N_2O emissions from a fine-loamy soil. Nutr Cycl Agroecosyst 59:177–191

Ruser R, Flessa H, Russow R, Schmidt G, Buegger F, Munch JC (2006) Emission of N_2O, N_2 and CO_2 from soil fertilized with nitrate: Effect of compaction, soil moisture and rewetting. Soil Biol Biochem 38:263–274

Ruser R, Fuß R, Andres M, Hegewald H, Kesenheimer K, Köbke S, Räbiger T, Suarez Quinones T, Augustin J, Christen O, Dittert K, Kage H, Lewandowski I, Prochnow A, Stichnothe H, Flessa H (2017) Nitrous oxide emissions from winter oilseed rape cultivation. Agri Ecosys Environ 249:57–69

Shaw LJ, Nicol GW, Smith Z, Fear J, Prosser JI, Baggs EM (2006) Nitrosospira spp. can produce nitrous oxide via a nitrifier denitrification pathway. Environ Microbiol 8:214–222

Severin M, Fuß R, Well R, Hähndel R, Van den Weghe H (2016) Greenhouse gas emissions after application of digestate: short-term effects of nitrification inhibitor and application technique effects. Arch Agron Soil Sci 62:1007–1020

Singh SN, Verma A (2007) The potential of nitrification inhibitors to manage the pollution effect of nitrogen fertilizers in agricultural and other soils: A review. Environ Pract 9:266–279

Stehfest E and Bouwman AL (2006) N_2O and NO emission from agricultural fields and soils under natural vegetation: summarizing available measurement data and modeling of global annual emissions. Nutr Cycl Agroecosyst 74:207–228

Sutton AL, Huber DM, Jones DD, Kelly DT, Bache DH (1986) Use of nitrification inhibitors and ammonia enrichment with swine manure applications. Appl Eng Agric 2:179–185

Volpi I, Laville P, Bonari E, Di Nasso N, Bosco S (2017) Improving the management of mineral fertilizers for nitrous oxide mitigation. The effect of nitrogen fertilizer type, urease and nitrification inhibitors in two different textured soils. Geoderma 307:181–188

Wolf U, Fuß R, Höppner F, Flessa H (2014) Contribution of N_2O and NH_3 to total greenhouse gas emission from fertilization. Results from a sandy soil fertilized with nitrate and biogas digestate with and without nitrification inhibitor. Nutr Cycl Agroecosyst 100:121–134

Wu D, Senbayram M, Well R, Brüggemann N, Pfeiffer B, Loick N, Stempfhuber B, Dittert K, Bol R (2017) Nitrification inhibitors mitigate N_2O emissions more effectively under straw-induced conditions favoring denitrification. Soil Biol Biochem 104:197–207

6 Effect of DMPP on direct N_2O emissions

6.1 Introduction

Several studies reported a mitigation of N_2O emissions from agricultural soils through nitrification inhibitors (NI) (*Halvorson* et al., 2010; *Venterea* et al., 2011; *Sanz-Cobena* et al., 2012; *Wu* et al., 2017). In a review from *Ruser & Schulz* (2015), the meta-analysis from *Akiyama* et al. (2010) was expanded by an actual dataset from 85 to 140. Herewith, the reduction potential of approximately 35 % of the N_2O emission reported by *Akiyama* et al. (2010) was confirmed. 3,4-Dimethylpyrazole phosphate (DMPP) is known as a highly effective NI (*Zerulla* et al., 2001a; *Weiske* et al., 2001) with non-toxicological or ecotoxicological side effects (*Zerulla* et al., 2001a; *Zerulla* et al. 2001b). The mitigation of N_2O emission by NIs is achieved by inhibiting the first step of nitrification, the oxidation of NH_3 by the enzyme ammonia monooxygenase (AMO) (*Arp & Stein*, 2003; Figure 6.1).
Beside this N_2O mitigation effect, NIs can have a positive effect on N use efficiency (NUE), thus decrease nitrate losses (*Ladha* et al., 2005; *Ruser & Schulz*, 2015; *Alonso-Ayuso* et al., 2016) and increase yield (*Pasda* et al., 2001). *Li* et al. (2008) found 6.6 % – 7.5 % higher oilseed rape grain yields in plots fertilized with urea + NI compared to plots with urea alone.

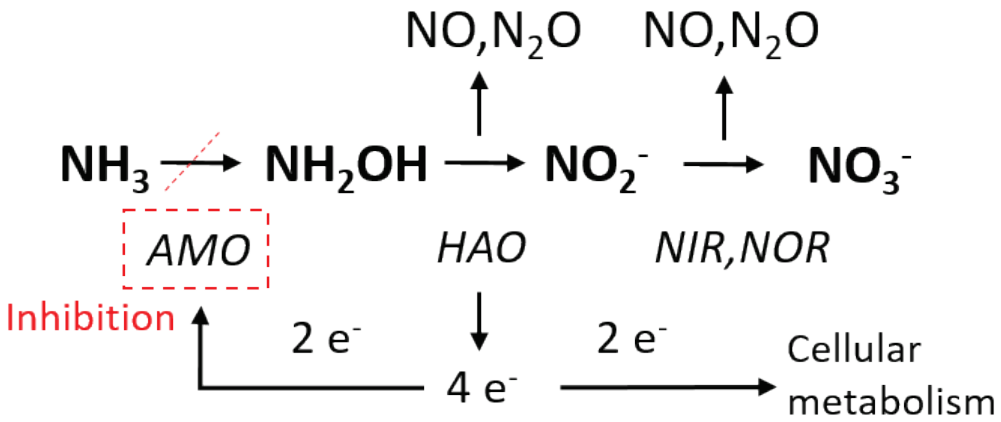

Fig. 6.1: Pathway of ammonia (NH_3) oxidation in NH_3 oxidizing bacteria (AOB), according to *Arp & Stein* (2003; modified). AMO = ammonia monooxygenase; HAO = hydroxylamine oxidoreductase; NIR = nitrite reductase; NOR = nitric oxide reductase.

In Chapter 5, the effect of the addition of NI to organic fertilizer at all five sites of the research project was described. To investigate the effect of NI added to mineral fertilizer in oilseed rape production, a further treatment was established in Hohenheim, using

the NI 3,4-dimethylpyrazole phosphate (DMPP) to evaluate the hypotheses:

(i) DMPP can significant reduce direct N_2O emissions
(ii) DMPP has a positive effect on OSR grain yield

6.2 Materials and Methods

The study was conducted at the site Ihinger Hof, Hohenheim described in detail in Chapter 3 and 4. The fertilizer ENTEC®26 (26 % N, 13 % S; NI= DMPP; *EuroChem Agro*) was used. Due to the fertilizer's sulphur content, no extra sulphur fertilization was applied to this treatment which is also in accordance with farmer's practice at Ihinger Hof. The ENTEC treatment was established simultaneously to all other treatments of the entire study (4 replicates). To evaluate the mitigation effect, the results from the treatment ENTEC were compared with those of N4 (Table 6.1). In contrast to the other treatments with mineral N fertilization, ENTEC was applied in one single dose. Due to the use of two fertilizer types, short-term effects of the different chemical specification could be possible.

Tab. 6.1: N-amount, application dose, nitrification inhibitor and fertilizer type used for the ENTEC experiment; abbreviations: DMPP= 3,4-dimethylpyrazole phosphate; ASN= ammonium sulphate nitrate; CAN= calcium ammonium nitrate.

Treatment	N amount kg N ha^{-1}	dose	NI	N fertilizer			
					N_{total}	NH_4-N	NO_3-N
					%		
ENTEC	180	single	DMPP	ASN	26	19	7
N4	180	split	none	CAN	27	13.5	13.5

N_2O was measured in the first experimental year from December to September, in the second year from OSR seeding date to September, and in the third year, from OSR seeding date until December to detect effects over the post-harvest period. Due to a very intensive measuring program, it was not possible to investigate treatment effects at later post-harvest periods in the first two years (Table 6.2). For trace gas flux determination, the same chamber type "Drössler" was used as descripted in Chapter 3 and 4.
Soil sampling, N_{min} analysis, water-filled pore space, bulk density, soil C and N analysis and yield as well as the weather records were carried out as described in Chapter 4 and 7.

Tab. 6.2: Period and duration of the N_2O measurement of the experiment.

Year	measuring period	days
1	11.12.12 - 24.09.13	287
2	03.09.13 - 16.09.14	371
3	23.09.14 - 29.12.15	462

Statistical tests for normality and homogeneity of variance were performed graphically for all data. Natural log-transformation (*Parkin & Robinson*, 1993) of N_2O and CO_2 emissions data was carried out prior to the analysis of variance. For the comparison of the cumulative N_2O emission an ANOVA was performed using the PROC MIXED procedure by SAS 9.4 (*SAS Institute*, 2016). For N_2O emissions over time, repeated measures ANOVA was performed using the PROC MIXED procedure, with an autoregressive AR(1) covariance structure to acknowledge for proximate correlation. The PROC MIXED procedure was used to test yield parameters.

6.3 Results and Discussion

Weather conditions

The weather within the investigation period was very irregular. The first year was characterized by long winter with a closed snow cover until March. The annual precipitation was 34 % higher than the long-term mean (Chapter 3, Table 3.3). The precipitation in the second year was also higher than the long-term mean. Contrarily, the third year was dry particularly during spring and summer. The annual mean temperature was predominately higher than the long-term mean for all three experimental years.

N_2O fluxes

Mean N_2O fluxes of the first year are shown in Figure 6.2a. Two distinct N_2O flux peaks were detected; one in N4 and in ENTEC before fertilization. Increased N_2O fluxes were measured immediately after fertilization in conjunction with precipitation. The highest mean flux in the first year was measured with 142 µg N_2O-N m^{-2} h^{-1} in treatment N4. The low soil moisture due the lack of precipitation between mid of June and end of July resulted in low N_2O fluxes (Figure 6.2a). With precipitation after harvest, the fluxes rose again.

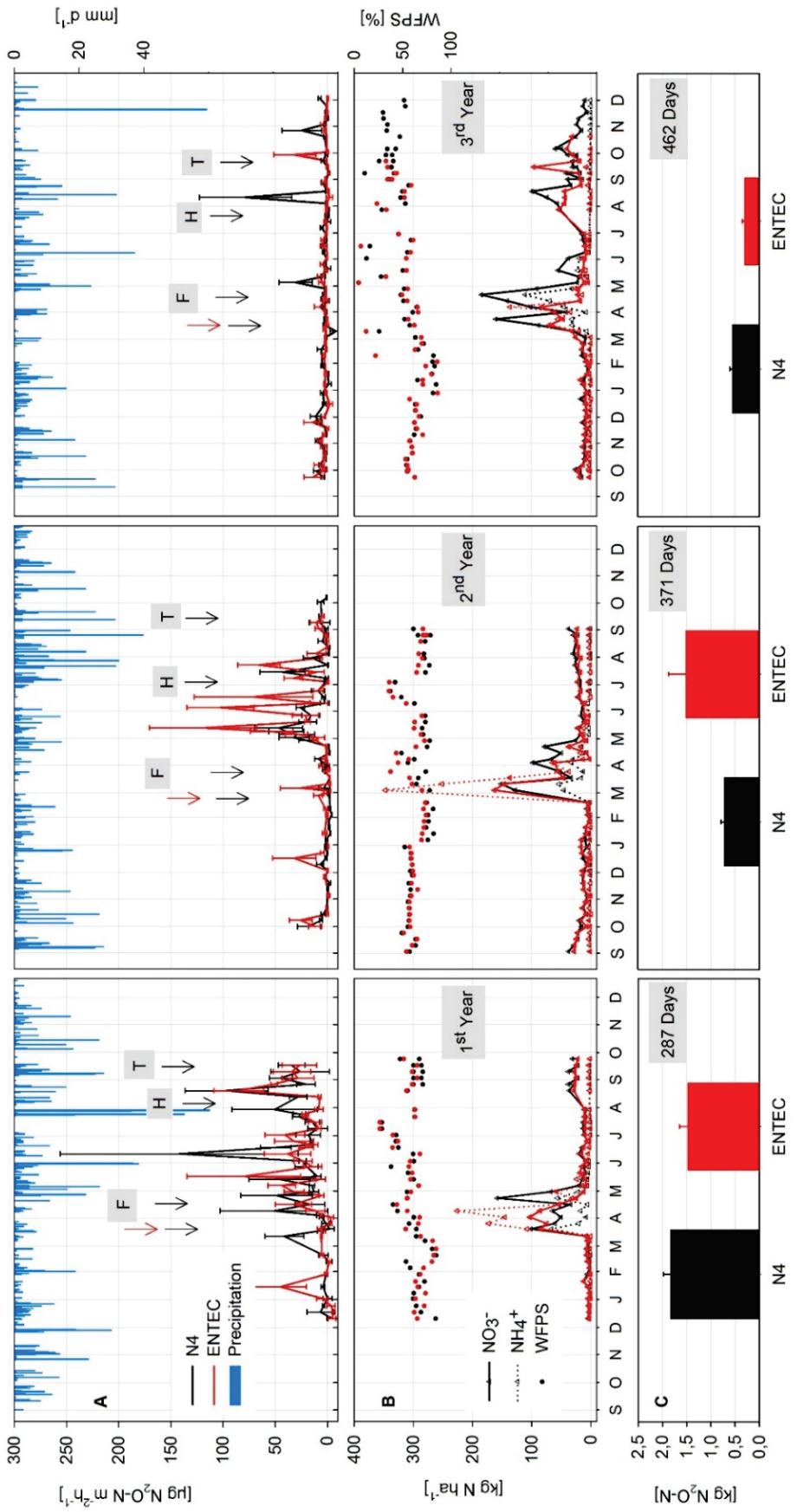

Fig. 6.2: Mean N_2O flux rates (n=4 ± standard error) and precipitation (a) and mean soil nitrogen and ammonium content with WFPS (b) from the treatment N4 and ENTEC in all three years; Mean cumulative N_2O emissions with standard error from treatment N4 and ENTEC (c); abbreviations: F= fertilization; H= harvest; T= tillage.

In the second year, the N_2O fluxes were low until fertilization and there was only a weak direct response to fertilization although soil moisture often exceeded 60 % WFPS, the threshold for denitrification (*Dobbie* et al., 1999; *Skiba & Ball*, 2002; *Batemann & Baggs*, 2005) (Figure 6.2b) and high mineral N concentrations in soil (Figure 6.2b). However, there was hardly any precipitation during the fertilization period. Approximately 4 weeks later, the N_2O fluxes increased strongly after some rainfall events. Also in the second year, the OSR harvest enhanced N_2O fluxes. The highest N_2O flux in the second year was measured in treatment ENTEC with 119 µg N_2O-N m^{-2} h^{-1}.

N_2O fluxes in the third year were low over the entire measurement period. There was hardly any response to fertilization or rewetting events. The only N_2O peak was measured after harvest paired with a precipitation (78 µg N_2O-N m^{-2} h^{-1}; N4; Figure 6.2a). From April, the WFPS threshold of 60 % was never reached again (Figure 6.2b).

Increased N_2O flux rates after fertilization paired with precipitation events were often observed (*Akiyama* et al., 2000; *Fuß* et al., 2011; *Yamamoto* et al., 2017). *Smith* et al. (1998) found a strong relationship between the magnitude of N_2O fluxes and the amount and distribution of rainfall during fertilization periods. After fertilization and rainfall events, high N_2O fluxes occur due to the increased availability of nitrate as substrate for N_2O production from denitrification together with anaerobic conditions (*Flessa* et al., 1995; *MacKenzie* et al., 1997). Many studies reported a threshold value of 60 % WFPS above which N_2O emissions strongly increased (*Dobbie* et al., 1999; *Skiba & Ball*, 2002; *Batemann & Baggs*, 2005). As we never reached this threshold value in the third year, low WFPS is probably the reason for the low fluxes measured in the third year given that the amount of nitrate as substrate for N_2O production was the same in every year.

After OSR harvest, soil nitrate contents were elevated in all three years (Figure 6.2b). Above the threshold of 10 mg nitrate N kg^{-1} soil, denitrification rates were shown to be independent on soil nitrate concentration (*Mosier* et al., 1983). This value was often reached after harvest during the entire investigation period, indicating that soil nitrate was not a limiting factor for denitrification. Consequently, C availability along with enhanced microbial activity with the related oxygen consumption were responsible for the increased fluxes after harvest.

Winters were mild in all three years. Therefore, considerable N_2O pulses during thawing of frozen soil as often observed in other investigations (*Flessa* et al., 1995; *Röver* et al., 1998; *Kaiser & Ruser*, 2000; *Ruser* et al., 2001) could not be expected.

The statistical model did not find significant effects of the nitrification inhibitor (ENTEC) on median daily flux rate in the first and third year (Table 6.3). In the second year, the mean daily flux was significantly higher ($p = 0.005$) in the treatment ENTEC compared to N4. There was a strong correlation ($p < 0.001$) between N_2O and CO_2 fluxes in all three years.

A correlation between CO_2 and N_2O fluxes indicates heterotrophic microbial denitrification as a main N_2O source during the investigated period. The C availability is a driving factor for denitrification (*Knowles,* 1982; *Beauchamp* et al., 1989), directly by increasing the energy

and electron donator to denitrifiers, and indirectly through enhanced microbial growth thereby stimulating high O_2 consumption (*Garcia-Montiel* et al., 2003, *Gillam* et al., 2008).
Other tested influencing factors (WFPS; NO_3^-; NH_4^+; temperature) for N_2O production showed no significant influence on daily fluxes (data not shown).

Tab. 6.3: Natural log back-transformed daily and cumulative N_2O emissions of the treatments N4 and ENTEC.

Year	N4		ENTEC		
	Median	SE	Median	SE	p value
ln back transformed daily N_2O flux [kg N_2O-N]					
1	10.1	0.63	11.3	0.70	0.614
2	1.6	0.11	4.1	0.31	0.005
3	2.8	0.06	2.2	0.04	0.384
ln back transformed cumulative N_2O emissions [kg N_2O-N]					
1	1.8	0.08	1.5	0.07	0.044
2	0.7	0.10	1.4	0.20	0.039
3	0.5	0.08	0.3	0.04	0.065

Cumulative N_2O emissions

Figure 6.2c shows the mean cumulative N_2O emissions in the three experimental years. Despite of the shortest measurement period of 287 days, the cumulative emissions were highest in the first year with 1.8 kg N ha^{-1} in the N4 treatment and 1.5 kg N ha^{-1} in the ENTEC treatment. There was a significant mitigation effect (17 %) in the ENTEC treatment (Table 6.3) in the first year. In the third year the emissions were low with no significant difference between the treatments. However, the mean emission for ENTEC was lower.
This is consistent with several other studies that reported a mitigation effect by the use of NI (*Akiyama* et al., 2010; *Pfab* et al., 2012; *Ruser & Schulz*, 2015). In the second year, the N_2O emissions from the ENTEC treatment were twice as high as the N4 treatment emissions with 1.4 kg N ha^{-1}. No immediate explanation could be found for that phenomenon. After the re-checking the entire process, possible errors related to fertilization, measurement, handling of samples, calculations, or large differences in soil water contents could be excluded.

OSR oil yield

In the first year, the OSR yield was 4.06 t ha^{-1} in the ENTEC treatment and 4.14 t ha^{-1} in treatment N4. This yield was significantly higher when compared to the following two years (Table 6.4). There was no effect of NI on yield. The average oil content was 47.6 % without differences between the years or treatments. Also, the oil yield was not affected by NI or year. The average oil yield was 18.9 hl ha^{-1}.
The OSR yield from this investigation was within the range given for the average OSR yields in Germany (3.90 – 4.48 t ha^{-1}) between 2013 and 2015 (*STATISTA*, 2017). *Li* et al. (2008) found 8 % higher OSR yields due to the use of the NI DMPP and other studies also reported a positive effect of NIs on yield (*Pasda* et al., 2001; *Liu* et al., 2013; *Martinez* et al., 2015). In

accordance to work published by other authors (*Weiske* et al., 2001; *Arregui & Quemada*, 2008; *Ercoli* et al., 2013), we could not observe increased yields due to NI in our investigation.

Tab. 6.4: Mean OSR yield, oil content and oil yield of the treatment ENTEC and N4 for all experimental years; different letters indicate significant differences between years (PROC MIXED; Tukey-test, $p < 0.05$).

Parameter	Year	ENTEC	N4
Yield	1	4.06 a	4.14 a
[t ha^{-1}]	2	3.86 b	3.87 b
	3	3.75 cb	3.00 cb
Oil content	1	47.4 a	47.4 a
[%]	2	47.8 a	47.4 a
	3	47.9 a	47.6 a
Oil yield	1	19.2 a	19.6 a
[hl ha^{-1}]	2	18.4 a	18.3 a
	3	18.0 a	18.9 a

6.4 Conclusion

Hypotheses one could only be confirmed in the first year with highest annual emissions during the entire investigation period. Here DMPP significantly reduced the direct N_2O emissions. NI´s can be an effective tool to mitigate N_2O emissions but the degree of utilization of the inhibitory potential is depending on site and weather conditions. In this investigation, NI did not increase OSR yield, therefore hypothesis two had to be rejected.

7 Nitrous oxide emissions after incorporation of winter oilseed rape (*Brassica napus* L.) residues under two different tillage treatments

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Abstract

To investigate the effect of crop residues from winter oilseed rape on N_2O emissions a field experiment in which crop residues of winter oilseed rape (*Brassica napus* L., OSR) were replaced with ^{15}N labelled OSR residues was conducted at a field experiment in South Germany, nearby Stuttgart. Nitrous oxide (N_2O) emissions and ^{15}N abundance in the N_2O were determined for a period of 11 months after harvest of OSR and in the succeeding crop winter wheat (*Triticum aestivum* L.). Measurements were carried out with the closed chamber method in a treatment with conventional tillage (CT) and in a treatment with reduced soil tillage (RT). High N_2O fluxes occurred in a short period after OSR residue replacement after harvest and after N-fertilization to winter wheat in the following spring. Cumulative N_2O emissions over the entire investigation period (299 days) ranged between 1.7 kg and 2.4 kg N_2O-N ha^{-1} with no significant treatment effects.
During the first eight weeks after OSR replacement more than half of the cumulative emissions occurred, highlighting the importance of this post-harvest period for annual N_2O

budgets of OSR. The contribution of residue N to the N_2O emission was low and explained by the high C/N-ratio fostering immobilization of mineral N. Only 0.03 % of the N_2O-N emitted in the conventional tillage treatment and 0.06 % in the reduced tillage treatment stemmed directly from the crop residues. The low contribution of the OSR residues to the direct N_2O emissions shows, that the current IPCC tier 1 approach, which assumes an EF of 1 %, strongly overestimated direct emissions from OSR crop residues.

8 Incubation study of winter oilseed rape residues

Results of the following Master Thesis are included in this Chapter:
"Einfluss von Rapsernterückständen auf umweltrelevante Nährstoffausträge in einem Mikrokosmenversuch", *Herr* (2015).

8.1 Introduction

Several studies reported a negative correlation between C/N–ratio and N_2O emissions (*Kaiser* et al., 1998; *Chen* et al., 2013). The incorporation of crop residues with low C/N–ratios often cause an increase in N_2O emissions (*Aulakh* et al., 2001; *Baggs* et al., 2000; *Wrage* et al., 2004). Most of these studies utilised crop residues from different plants to investigate the effect of low and high C/N–ratios. Depending on plant species, quality traits like polyphenol content, lignin content or protein-binding capacity differ and also their influence on N_2O release. But within a plant species, C/N–ratios can vary, depending on the N fertilization. In a microcosm trial, *Pfab* (2011) investigated the effect of broccoli crop residues with different C/N–ratios on N_2O emissions from a silty soil. The C/N–values ranged from 8.2 and 17.1 due to different N fertilization. *Pfab* (2011) found no significant differences between the treatments for cumulative N_2O emissions but a tendency for higher emissions in treatments with low C/N crop residues (Figure 8.1). *Pfab* (2011) explained that with an enhanced mineralization and nitrification connected with an increased turnover of organic matter with high CO_2 production and oxygen depletion, favouring denitrification.
The level of N fertilization has not just an effect on C/N–ratios of OSR crop residues, also the accruing amount of biomass is affected. *Velthoff* et al. (2002) reported about an enormous increase of N_2O emissions from soil amended with elevated amounts of crop residues from sugar beet. Also *Pfab* (2011) found significantly increased N_2O emissions with increasing amounts of crop residues (Figure 8.1). *Pfab* (2011) explained the high N_2O emissions with the increased amount of easily available carbon due to the high amount of crop residues. Through this, the turnover of organic matter is accelerated with the same effect of favouring denitrification as described previously, depending on the C/N–ratios of the residues. For crop residues with C/N–ratios above 25, *Smith & Smith* (2009) reported about a positiv correlation between the amount of crop residues and net NO_3^- immobilization. Such a high immobilization can have a mitigation effect on N_2O emissions.

The amount and quality of crop residues can also influence NO_3^- leaching. Leaching losses are higher with increased amount of crop residues with low C/N–ratios because of enhanced mineralization and nitrification in contrast to crop residues with C/N–ratios above 25, which can induce NO_3^- immobilization. *Pfab* (2011) reported about significant higher NO_3 losses from treatments with low C/N–ratios whereas an effect of the amount of residue input was not detectable (Figure 8.2).

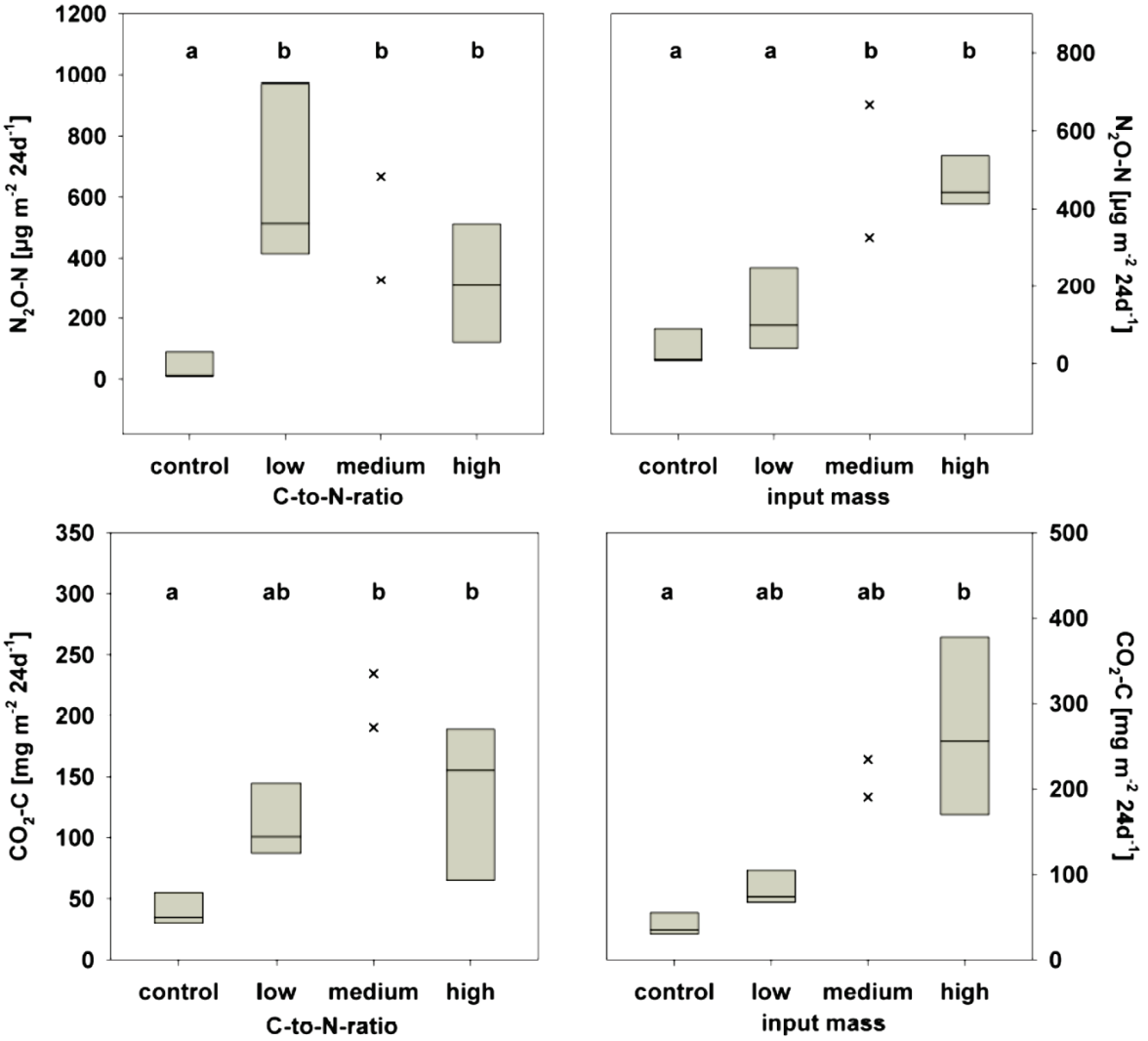

Fig. 8.1: Median cumulative N_2O-N and CO_2-C emissions (N=4) measured in a microcosm system during 24 days after addition of cauliflower residues (*Pfab*, 2011). Different superscript letters indicate statistically significant differences between groups (Student-Newman-Keuls test, $p < 0.05$).

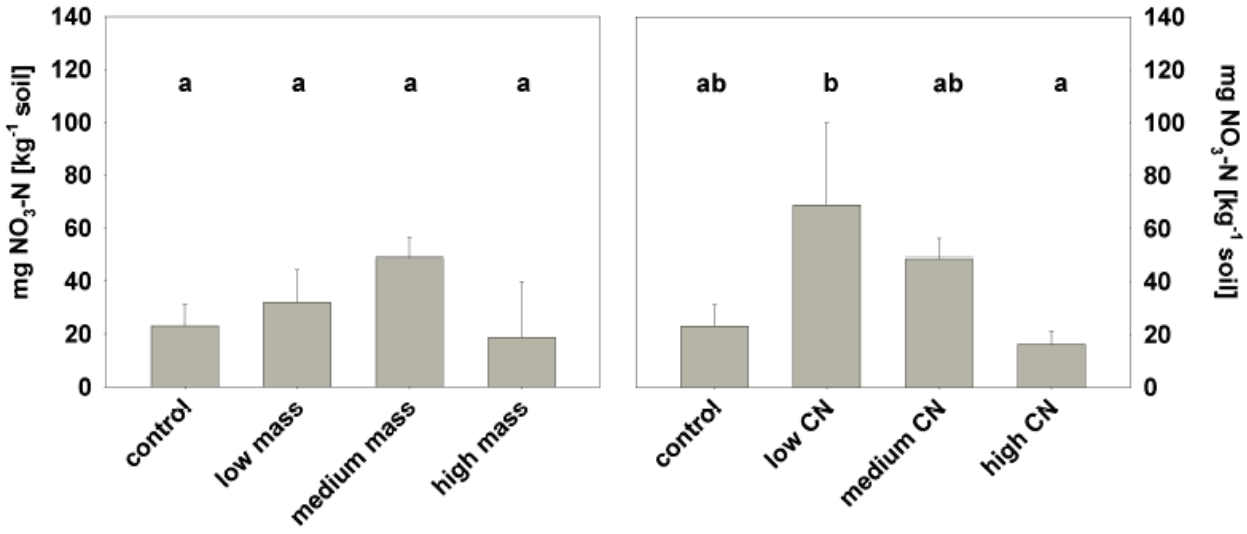

Fig. 8.2: Results from *Pfab* (2011); left: Mean nitrate-N content per kg soil in the control and in the treatment with low, medium and high amount of the residue input. Right: mean nitrate-N content per kg soil in the control and in the treatment with low, medium and high C/N–ratio of applied residues. Different superscript letters indicate statistically significant differences between groups (Student-Newman-Keuls Test, $p < 0.05$).

The aim of the study was to investigate the effect of the amount and quality from OSR crop residues on N_2O emissions and NO_3^- losses at the site Ihinger Hof (research station, Hohenheim) as complement to Chapter 7. To evaluate these effects, a microcosm system was established to determine following hypotheses:

(i) OSR crop residues with low C/N–ratios cause higher N_2O emissions
(ii) N_2O emissions increase with increasing amounts of OSR crop residues
(iii) NO_3^- leaching increases with increasing OSR crop residue amounts and narrow C/N–ratios

8.2 Materials and Methods

8.2.1 Experimental setup

Preparation

Soil was taken from the uppermost horizon (0-30 cm) of the field described in Chapter 3 and 4, air dried and sieved (5 mm). Plexiglas cylinder (height 30 cm, inner diameter 14.4 cm) were packed up to a height of 15 cm with soil compacted to a bulk density of 1.2 g cm^{-3}, corresponding to 2.388 g soil for each column. At the bottom of the columns, cellulose acetate membrane filters (0.45 µm) were inserted to prevent blockage effects. An opening with an airtight tube on the bottom lid allowed to collect percolate of the column. The system was drained by a pump (-100 mbar). The columns were sealed airtight by a removal top lid with

a screw cap for irrigation. The headspace of the columns was continuously flushed with atmospheric air (20 ml min^{-1}) in the microcosm system. The microcosm system was placed in a dark climate chamber with constant temperature of 20°C.
After packing the columns and assembly of the microcosm system, the soil was irrigated for ten weeks with 0.025 M $CaCl_2$ (30-60 ml d^{-1}) to rewet the soil and reactivate microbial activity as well as to leach surplus NO_3^--N. $CaCl_2$ solution was chosen to maintain the aggregate stability. After reaching the threshold of ≤ 100 mg NO_3^- L^{-1} in the percolate irrigation was terminated.

Crop residues

The used OSR crop residues were collected from the N-dosing test at the Ihinger Hof 2014, described in Chapter 4. The amount and C/N–ratios of the crop residue are shown in Figure 8.3. The treatments of this microcosm study were based on these results (Table 8.1). Crop residues were dried, ground and mixed into the uppermost 7.5 cm of the soil column. Control columns were treated in the same manner but without addition of residues. After the treatment, the columns were taken back into the microcosm system and arranged in a randomized complete block design.

Gas sampling

For gas sampling, 22.4 ml vials with a crimp-cap septum (*Häberle, Germany*) were connected with the exhaust hose of the column headspace using a cannula. To avoid overpressure in the vials, a second cannula was pricked in the septum which enabled flushing of the vial. Furthermore, 4 samples were taken from the incoming air during every sampling event. In the first 30 days, gas samples were taken twice a day, afterwards once a day. First measurement was done after crop residue incorporation. N_2O and CO_2 composition were analysed using a gas chromatograph equipped with a ^{63}Ni electron capture detector (ECD) (*450 Bruker*) and autosampler (*GX-281 Gilson*).

Irrigation system

From the beginning until day 30, irrigation was done manually with a 0.025 $CaCl_2$ solution twice a day (morning and evening) and afterwards daily (morning). In the first 17 days, the irrigation amount was 15 ml $microcosm^{-1}$, then 30 ml corresponding to the mean daily precipitation of the site Ihinger Hof.

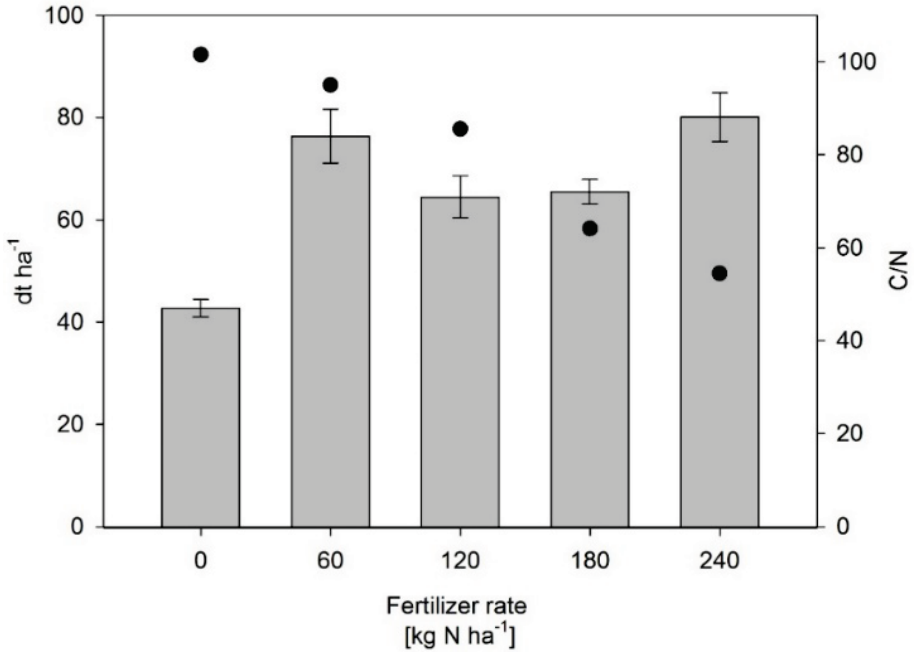

Fig. 8.3: Mean quantities of OSR crop residues and mean C/N–ratios affected by N fertilization (n=4, ± SE) of the field study 2014, described in Chapter 4 (site Hohenheim). Dots: C/N–ratio; bars: amount of crop residues (n=4, ± SE).

Tab. 8.1: Characteristics of the established treatments with OSR residue C/N–ratios and applied rate per microcosm.

treatment	name	C/N-ratio	application rate	C/N	application rate g (microcosm)$^{-1}$
1	control	control	-	-	0
2	low CN	low	high	54,5	10,6
3	medium CN high input	medium	high	85,5	10,6
4	high CN	high	high	101,5	10,6
5	medium CN low input	medium	low	85,5	5,3

Soil and percolate analyses

Soil samples were taken at the beginning and at the end of the study to determine the mineral N content. For quantification, 20 g of fresh soil were extracted with 80 ml of a 0.0125 M $CaCl_2$ solution. Percolate was collected once a week. For both soil extract and percolate, a flow analyzer (3 *QuAAtro, SEAL Analytical, UK*) was used to measure the concentration of nitrate (NO_3^-) and ammonium (NH_4^+). Soil moisture was determined gravimetrically after drying an aliquot of fresh soil at 105°C for 24 h. The calculation of the water-filled pore space (WFPS) was given in detail by *Ruser* et al. (1998).

8.2.2 Calculation and statistics

N_2O-N and CO_2-C fluxes per microcosm were calculated using the following equation:

$$\text{Flux rate } (m^{-2}\ h^{-1}) = \frac{((C_s - C_a) \cdot k \cdot \frac{273.15}{T} \cdot F}{A}$$

c_s = concentration in the sample (ppb)
c_a = concentration in the atmospheric air (ppb)
k = conversion factor (N_2O: 1.25 µg N_2O-N μL^{-1}; CO_2: 0.536 µg CO_2-C μL^{-1})
T = Temperature in the microcosm system (K)
F = Flow rate (L h^{-1})
A = microcosm surface (m^2)

Due to technical problems, 7 of the 21 microcosms have been eliminated from the evaluation. In these columns the soil water contents were too high because of a defective negative pressure system.
Statistical analyses were conducted using R (version 3.1.3, *R core*, 2015). Natural log-transformation of the N_2O emissions data was carried out prior to the analysis. Variance homogeneity of cumulative emissions were tested by a Bartlett-Test. Differences between the treatments were tested using a One Way Anova, significant differences were determined using Student–Newman–Keuls Test ($\alpha = 0.05$).

8.3 Results and Discussion

Directly after the addition of crop residues, the N_2O flux rose in the treatment "medium CN low input" (Figure 8.4A). The highest flux after initiation of the experiment was measured with 187 µg N_2O-N m^{-2} h^{-1} in the control without crop residues. In the following days the fluxes dropped down in all treatments to a low level. Only the control fluxes rose up again after day 10. A peak was measured after doubling the irrigation amount at day 17 in all treatments except "high CN" and in the control. In this period the highest peak during the entire experimental period was detected in the treatment "medium CN low input" with 440 µg N_2O-N m^{-2} h^{-1}. Except for the control treatment, all treatments dropped down again after this peak. From day 30 all treatments had low fluxes with a slight rise in treatment "high CN". There were no significant differences between the treatments for cumulative N_2O emission (Figure 8.4C). Treatments with crop residues showed a trend to lower emissions compared to the control.

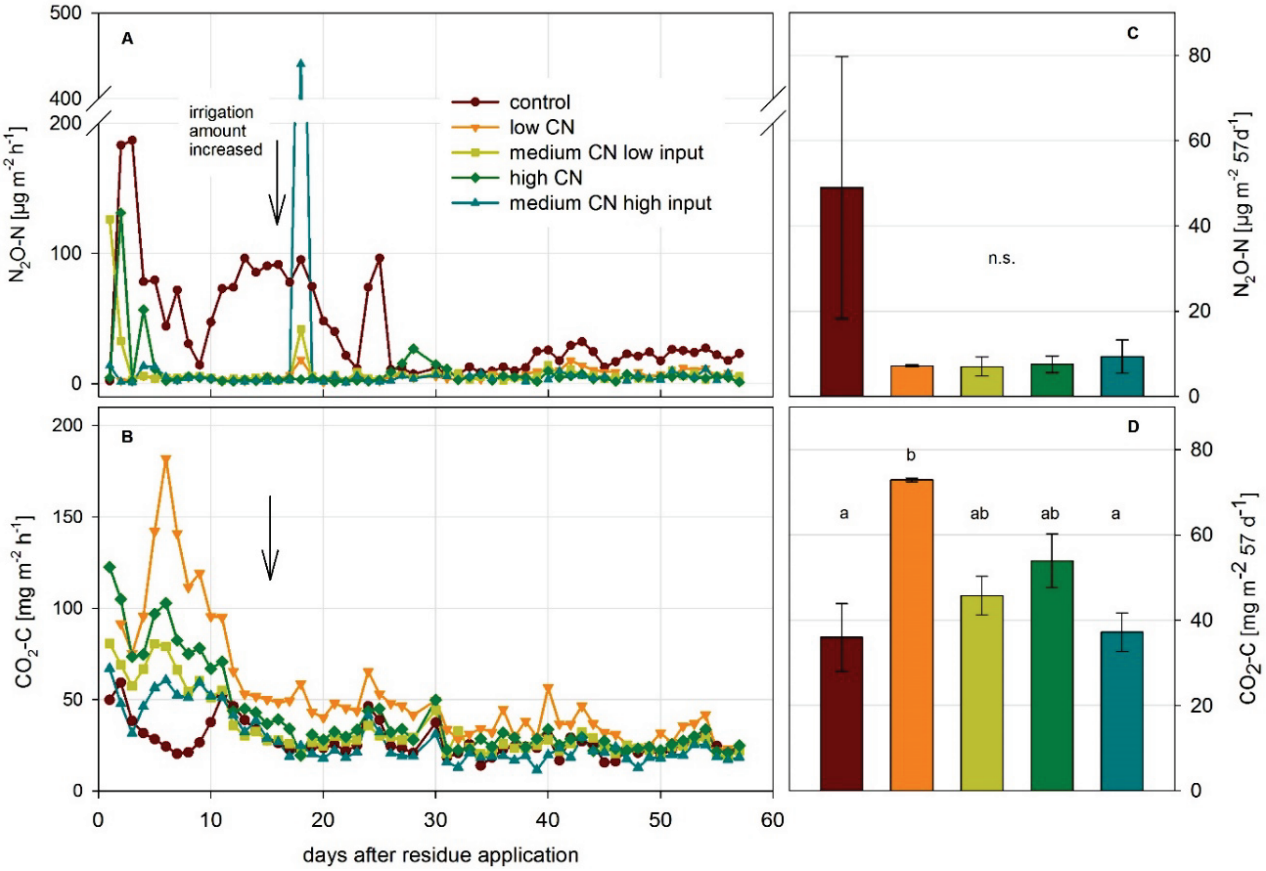

Fig. 8.4: A, B = Mean daily N_2O-N and CO_2-C fluxes measured in the microcosm system during 57 days after crop residue application effected by C/N- ratio and input mass; C, D = Mean cumulative N_2O-N and CO_2-C emissions (± SE) during the experimental period. Different letters indicate statistically differences (Student-Newman-Keuls test, $p < 0.05$).

Corp residue application provides easily available labile carbon and nitrogen as nutrient substrates, which in turn can increase the microbial activity in the soil. Resulting rapid oxygen consumption by microbes during respiration decreases the redox potential and thus favors conditions for denitrification (*Flessa & Beese*, 1995; *Azam* et al., 2002; *Miller* et al., 2008). In this incubation experiment, we could not observe such an effect. Although the fluxes rose in all treatments, the control without crop residue application had the highest fluxes. It is likely that the incorporated crop residues led to a fast NO_3^- immobilization due to the high C/N–values between 55 and 102 (*Powlson* et al., 2001; *Smith & Smith*, 2009), whereby it was unavailable as substrate for denitrification. *Chaves* et al. (2007) observed a low share of plant available NO_3^-- N in soil after incorporation of organic materials with high C/N–ratios. *Kaewpradit* et al. (2008) observed a reduction of N_2O emissions after incorporation of organic materials with high C/N–ratios.

It is highly probable that also in this incubation experiment N was the limiting factor in treatments amended with crop residues. Another reason for the low fluxes could be the low soil water content until day 17. The column irrigation was stopped two weeks before crop

residue application to ensure a good incorporation. Due to the high O_2 partial pressure after crop residue application, the conditions for denitrification were unfavourable whereas the conditions for nitrification were optimal, with marginal loss of N_2O (*Arp & Stein*, 2003).
The higher fluxes of the control could be explained by the enhanced microbial activity and mineralization in the absence of NO_3^- immobilization after mixing and compaction at day 0 (*Gregorich* et al., 1989).
The mean CO_2 fluxes showed a different time course compared to the N_2O fluxes (Figure 8.4B). The highest peak was measured after 6 days with 182 mg CO_2-C m^{-2} h^{-1} in treatment "low CN". This treatment emitted the highest fluxes during the entire experimental period. In the first 10 days, the lowest fluxes were measured in the control. There was hardly no response to doubling irrigation at day 17. From that day onwards, fluxes in the treatments maintained a similarly low course and fluctuated from 11 to 65 mg CO_2-C m^{-2} h^{-1}. The cumulative CO_2 emissions are shown in Figure 8.4D. The control and the treatment "medium CN low input" had significantly lower emissions compared to treatment "low CN" with 73 mg CO_2-C m^{-2} 57 d^{-1}. The treatments "medium CN high input" as well as "high CN" did not show significant difference to the other treatments. CO_2 is a product of several dissimilar processes and an indicator for microbial activity (*Bartling* et al., 2009; *Sölter & Weber*, 2000). The slight delay of CO_2 fluxes may have been caused inter alia by the initial growth of microbes as well as enzyme syntheses (*Bartling* et al., 2009).
At the beginning of the experiment, there were no differences in the mineral N contents of the columns (Table 8.2). After the experiment, the mineral N contents were lower in all treatments. With 1.3 mg N kg^{-1}, the control had significantly higher values than all other treatments. There was no effect of C/N–ratio or input amount on mineral N content. The total loss of nitrate by percolate ranged between 10.8 and 19.9 mg microcosm^{-1} and was not affected by crop residues. There was no significant difference between the treatments. The treatments "medium CN high input" and "high CN" showed a trend to lower losses.
The reduction of mineral N after 57 days could be an evidence of N immobilization due to the high C/N–ratios of the crop residues. In support of this argument, the mineral N content of the control was the highest, where no immobilization occurred. Comparable results were reported by *Goyal* et al. (1992) und *Garcia-Ruiz & Baggs* (2007). For the same reasons the control as well as the treatments with low C/N crop residues likely showed a trend to higher nitrate content in the percolate.

Tab. 8.2: Mean N_{min} (± SE) content per kg soil at Day 0 and after the experiment; Nitrate N (NO_3^--N) leaching by percolate (mg microcosm^{-1}). Different letters indicate statistically significant differences (Student-Newman-Keuls test, $p < 0.05$).

Treatment	Soil				Percolate	
	N_{min}				Percolate NO_3^--N	
	Day 0		Day 57			
	[mg kg^{-1}]				[mg MC^{-1}]	
control	8.9a	± 0.0	1.3a	± 0.0	19.0a	± 6.4
low CN	8.1a	± 0.1	0.9b	± 0.1	17.5a	± 1.7
medium CN high input	7.2a	± 0.8	0.9b	± 0.0	10.8a	± 1.6
high CN	7.9a	± 1.0	1.0b	± 0.0	11.9a	± 2.7
medium CN low input	9.2a	± 0.6	1.0b	± 0.1	19.9a	± 2.7

8.4 Conclusion

It could not be confirmed that the OSR crop residues with low C/N–ratios cause higher N_2O emissions. There were no significant differences and no trends between the treatments. Also, the amount of crop residues did not show any effect on the emissions. The mineral N content in soil indicated N immobilization due to the crop residues. This might presumably be the main reason for the low N_2O fluxes. The general opinion of high N balances due to OSR crop residues could not be confirmed in this experiment. In contrast, OSR crop residues could temporary inhibit N losses after incorporation. The results of this incubation experiment correspond to the results of the field trial in Chapter 7.

9 Effect of tillage on N_2O emissions

9.1 Introduction

In comparison to conventional tillage systems (CT) that require a plough, reduced tillage (RT) is distinguished by elimination of soil inversion, reduction of soil disturbance by restricting mechanical interventions to shallow depths, and conservation and management of crop residues (*Cunningham* et al., 2004). RT practice is popular in Europe because of low crop production costs and reduced soil compaction (*Holland*, 2004). About 10–12 % of arable land (16 Mha) in Europe is under reduced tillage systems, including non-inversion tillage, eco-tillage, minimum tillage, mulch tillage, reduced tillage, zone tillage or no-tillage (*Abdalla* et al., 2013). In this investigation, RT is defined as tillage without soil inversion by the use of a chisel plough to 15 cm depth.
RT can increase C sequestration in the uppermost soil layer (*Alvarez*, 2005) and reduce soil erosion by an increased infiltration (*Lampurlanés* et al., 2001; *Copec* et al., 2015) due to improved soil pore structure and stability (*Oades*, 1984). Results of previous studies about the impact of RT on N_2O emissions were inconsistent. Several studies reported higher N_2O emissions under RT compared to CT associated with increased rates of denitrification, greater bulk density and soil water content (*Palma* et al., 1997; *Goossens* et al., 2001; *Ball* et al., 2008; *D'Haene* et al., 2008; *Abdalla* et al., 2013; *Lognoul et* al., 2017). Some studies reported lower (*Gregorich* et al., 2006; *Chatskikh & Olesen*, 2007; *Mutegi* et al., 2010; *Koga*, 2013; *Wang & Dalal*, 2015) or similar N_2O emissions to CT (*Chatskikh* et al., 2008; *Liu* et al., 2006; *Abdalla* et al., 2010; *Negassa* et al., 2015).

In this experiment, the effect of two different tillage systems on N_2O emission and OSR grain yield were investigated, with the following hypotheses:

(i) N_2O emissions from reduced tillage system are significantly higher than from a ploughed system.
(ii) OSR grain yield is not affected by tillage practise

9.2 Materials and Methods

The study was conducted at the site Ihinger Hof, Hohenheim described in Chapter 3. The treatment RT was established parallel to all other treatments (4 replicates each). Treatment N4, used as the conventional tillage system (Table 9.1), was compared with a RT treatment. To simplify, treatment N4 is called in this investigation CT, for conventional tillage (Table 9.1). N-fertilizer was applied simultaneously to all other treatments of the overall experiment. For soil cultivation before sowing, a chisel plough (approx.15 cm) was used for treatment RT

and a mouldboard plough (approx. 30 cm) for treatment CT. The investigation periods are shown in Table 9.2.

Tab. 9.1: Treatments of the experiment: N-amount, and fertilizer type; abbreviations: RT= reduced tillage; CT= conventional tillage; CAN= calcium ammonium nitrate.

Treatment	N amount kg N ha^{-1}	N fertilizer
RT	180	CAN
CT	180	CAN

Tab. 9.2: Period and duration of the N_2O measurement of the soil tillage experiment.

Year	measuring period	days
1	11.12.12 - 03.09.13	266
2	01.10.13 - 16.09.14	350
3	23.09.14 - 06.10.15	378

N_2O fluxes were measured in the first year between the initiation of the investigation in December to September in the second year, and in the following two years from OSR seeding to OSR harvest. Chamber type "Drössler" was used as described in Chapter 4. Soil sampling, N_{min} analysis, water-filled pore space, bulk density, C and N analysis of the soil and yield as well as the weather records were carried out as described in Chapter 4.
Statistical tests for normality and homogeneity of variance were performed graphically. Natural log-transformation (*Parkin & Robinson*, 1993) of the N_2O emissions data was carried out prior to the analysis of variance. For the comparison of the cumulative N_2O emissions an ANOVA was performed using the PROC MIXED procedure using SAS 9.4 (*SAS Institute*, 2016). For N_2O emissions over time, repeated measures ANOVA was performed using the PROC MIXED procedure, with an autoregressive AR(1) covariance structure to acknowledge for proximate correlation. Yield parameters were tested by using the PROC MIXED procedure.

9.3 Results and Discussion

Weather conditions

As described before, the weather within the investigation period was very irregular. The first year was characterized by intensive winter with a snow cover until March. The annual precipitation was 34 % higher than the long-term mean (Chapter 4, Table 1). In the second year, the precipitation was also higher than the long-term mean. In contrast, the third year was dry, particularly in spring and summer. The annual mean temperature was predominately higher than the long-term mean for all three experimental years.

N_2O fluxes

In the first year N_2O fluxes increased after fertilization, in conjunction with precipitation (Figure 9.1A). In this period, the highest flux of 210 µg N_2O-N m^{-2} h^{-1} was measured in treatment RT. Between mid of June and end of July, the fluxes were low, corresponding to a very low soil moisture (Figure 9.1B) due to the lack of precipitation. After a heavy rainfall events with 52, 36 and 60 ml d^{-1}, the fluxes rose again concurrently to OSR harvest at the end of July. In the second year, the level of N_2O fluxes was overall low when compared to the first year (Figure 9.1A). There was hardly no response to N-fertilization. Despite high N_{min} contents, fluxes remained low due to the low precipitation and corresponding well aeration of the topsoil (Figure 9.1C). At the end of April, rewetting of dry soil after strong rainfall events induced increased N_2O fluxes until soil moisture decreased again. After OSR harvest, a small N_2O emission peak occurred in both treatments. The highest flux with 46 µg N_2O-N m^{-2} h^{-1} was measured in treatment CT. N_2O fluxes were lowest in the third year, just three peaks were detected during the entire period (Figure 9.1A). After February, the WFPS was low most of the time (Figure 9.1B). The first two peaks were measured after N-fertilization, the third and highest peak with 60 µg N_2O-N m^{-2} h^{-1} was measured after harvest and rainfall in August.

Several studies reported increased fluxes after fertilization in conjunction with precipitation (*Akiyama* et al., 2000; *Fuß* et al., 2011; *Yamamoto* et al., 2017). After rainfall, high N_2O fluxes can occur due to the increased availability of nitrate as substrate for N_2O production alongside anaerobic conditions by high WFPS content in soil (*Flessa* et al., 1995; *MacKenzie* et al., 1997). Above 10 mg nitrate N kg^{-1} soil, denitrification rates are independent of soil nitrate level because of the saturation of the nitrate converting enzymes (*Mosier* et al., 1983). This value was often reached during the investigation period; soil nitrate was therefore not a limiting factor for denitrification. Due to mild winters, no considerable N_2O pulses during thawing of frozen soil were measured, as often observed in other investigations (*Flessa* et al., 1995; *Röver* et al., 1998; *Kaiser & Ruser*, 2000; *Ruser* et al., 2001).

In the first year, the median daily N_2O flux was significantly higher in the CT treatment than in RT (Table 9.4). There was a significant effect of CO_2 emission (Table 9.3). With 13.1 µg N_2O-N and 7.9 µg N_2O-N m^{-2} h^{-1}, the daily N_2O fluxes in the first and second year were 83 % and 36 % higher in comparison to the third year, respectively (Table 9.4). The fluxes in the second year were equal and considerably lower than in the first year and influenced by NO_3 and CO_2. In the third year, despite of the very low fluxes, the daily N_2O fluxes were significantly higher in the RT treatment than in CT. As in previous years, N_2O fluxes correlated significantly with CO_2 fluxes as an indicator for heterotrophic microbial denitrification as the main N_2O source.

Tab. 9.3: Natural log back transformed daily flux and cumulative N_2O emissions of the treatments CT and RT.

Year	CT		RT		
	Median	SE	Median	SE	p value
daily N_2O flux [µg N_2O-N m^{-2} h^{-1}]					
1	13.1	0.19	7.9	0.20	**0.038**
2	6.6	0.07	6.5	0.07	0.930
3	2.2	0.07	5.1	0.07	**0.001**
cumulative N_2O emissions [kg N_2O-N]					
1	2.2	0.11	1.7	0.16	0.434
2	0.8	0.02	0.9	0.02	0.879
3	0.4	0.17	0.5	0.20	0.653

Tab. 9.4: Type 3 test of fixed effects on daily fluxes of all three years; abbreviation: T = tillage.

	1st Year	2nd Year	3th Year
Effect		Pr > F	
T	**0.038**	0.930	**0.001**
Block	**0.025**	**0.003**	**0.014**
WFPS	0.970	0.562	**0.002**
NO_3	0.088	**0.041**	0.896
CO_2	**0.023**	**<.0001**	**<.0001**

Cumulative N_2O emissions

Mean cumulative N_2O emissions are shown in Figure 9.1D. The highest emissions were measured in the first year with 2.2 (CT) and 2.0 (RT) kg N ha^{-1}. In the following two years, the emissions dropped. There was no tillage effect on cumulative N_2O emissions in any of the three years (Table 9.4).

Similar emissions from CT and RT contradict the findings of some investigations (*McKenzie* et al., 1997; *Baags* et al. 2003), but nevertheless, are in agreement with those of other studies (*Liu* et al., 2006; *Negassa* et al., 2015). *Mahli & Lemke* (2007) found in three years of a 4-yr rotation cycle no differences between RT and CT tillage in cumulative N_2O emissions.

Higher N_2O fluxes under reduced tillage were often observed (e.g. *Johnson* et al., 2005; *Venterea* et al., 2005), particularly in the first years after changeover from CT to RT (*D'Haene* et al., 2008). Reason for the high emissions was the high water-filled pore space, which favoured denitrification. Also in our experiment, the WFPS of RT was generally higher than that of CT, also due to the higher bulk density and the resulting lower pore volume (Table 3.2). Nevertheless, the N_2O emissions were not affected by tillage.

OSR oil yield effected by tillage

OSR yield, oil content and oil yield were not affected by tillage or year (Table 9.5). This agrees with results from *Paré* et al. (2015). They reported from a 12-year study, that soil tillage did not affect OSR grain yield during 63 % of the experimental period (7–8 years out of 12), whereas yield and oil yield in the RT treatment in the remaining years did not show any consistent trend. This was in agreement with results reported from *Gruber* et al. (2012),

who did not find different OSR yield between RT and CT treatment in a 10-year tillage experiment.

Tab. 9.5: Mean OSR yield (n=4), oil content and oil yield of the treatment CT and RT of all experimental years. There were no significant effect of tillage or year (PROC MIXED; Tukey-test, $p < 0.05$).

Parameter	Year	CT	RT
Yield	1	4.14	3.78
[t ha^{-1}]	2	3.87	3.97
	3	3.90	4.06
Oil content	1	47.4	47.0
[%]	2	47.4	47.6
	3	47.6	47.8
Oil yield	1	19.6	19.6
[hl ha^{-1}]	2	18.3	18.9
	3	18.9	19.4

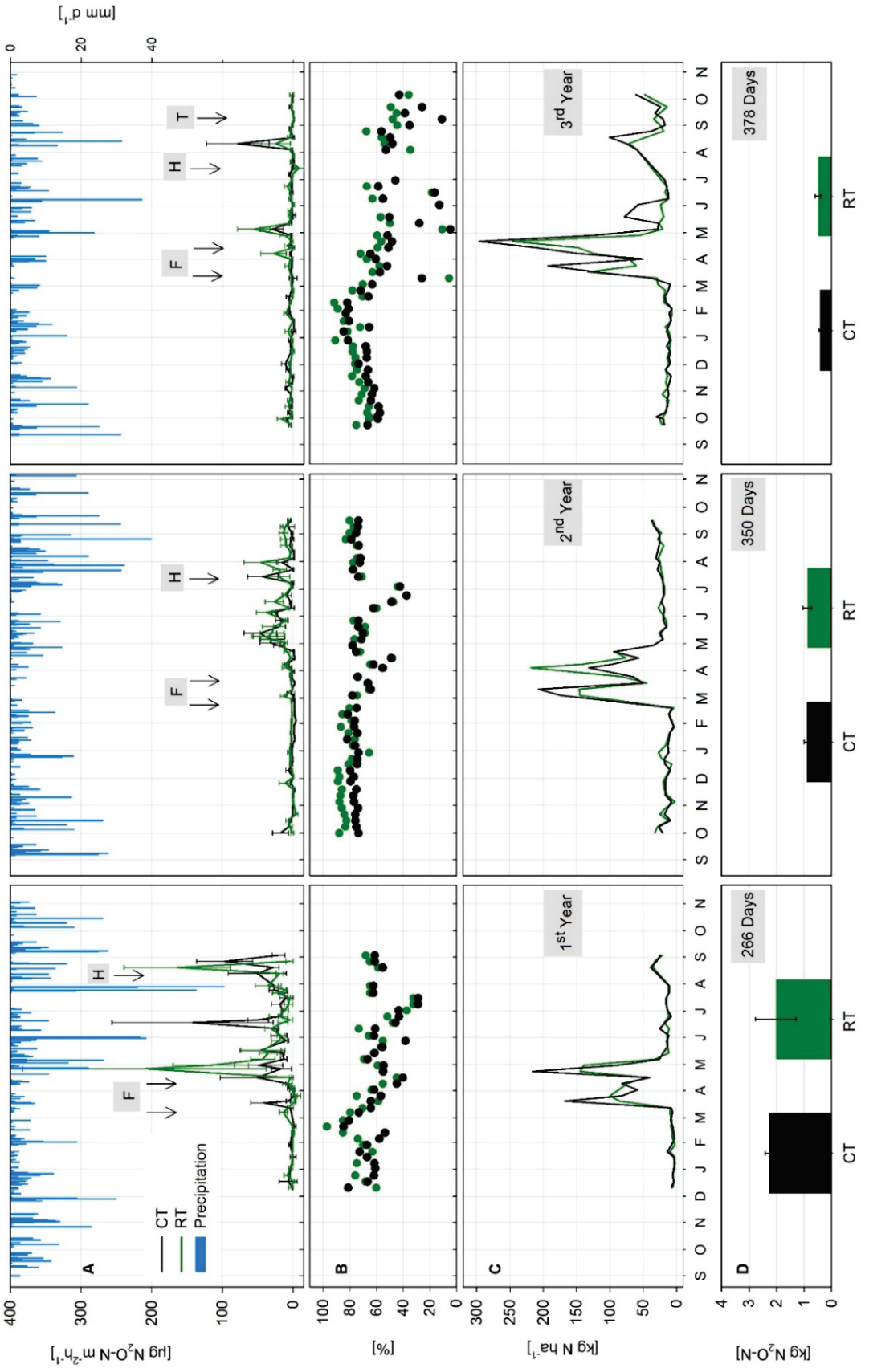

Fig. 9.1: Mean N_2O fluxes (n=4 ± SE) and daily precipitation (A); water filled pore-space (B), soil mineral N (C) and the cumulative N_2O emissions of the treatment conventional tillage (CT) and reduced tillage (RT) of all three year; abbreviations: F= fertilization; H= harvest.

9.4 Conclusion

Soil tillage did not affect N_2O emissions in this investigation, hypothesis one could not be confirmed. A possible reason for that could be the general low precipitation and rainfall distribution during the investigation period, particularly in the third year. In the entire experiment the fluxes were low in all investigated treatments at the site Hohenheim were generally low. Tillage did not affect OSR yield, therefore hypotheses two could be confirmed. In consideration of the lower operating costs using a chisel plough (RT) compared to a mouldboard plough, reduced tillage could be a better alternative, whereas possible higher costs for crop protection have also to be taken into consideration.

10 General Discussion

Effect of N fertilization on N_2O emissions from OSR

With this study on N_2O emissions from OSR we enlarged the dataset of Walter et al. (2015) by 50 %. Following the same methodology, we derived an exponential model relating N_2O emissions to N fertilization. The model confirmed a strong impact of study sites and years on annual N_2O fluxes. A nonlinear response of N_2O emissions to N fertilization has often been reported and explained either with an increased N supply strongly exceeding N demand of the crop or with extended periods of increased mineral N supply for N_2O production (van Groenigen et al., 2010; Hoben et al., 2011; Shcherbak et al., 2014). The global fertilizer related emission (FRE) factor derived from the exponential model was 0.6 % (fertilizer amount 200 kg N ha^{-1} a^{-1}). This factor was within the uncertainty range of the EF1 IPCC emission factor (0.3 % – 3 %), but about 40 % lower than the IPCC default value and was it also lower than the FRE factor calculated by GNOC (Global Nitrous Oxide Calculator, Edwards et al., 2013) and by Walter et al. (2015). Two of our five study sites have sandy soil texture with low Corg contents. The low water-holding capacity of these sites results in well aeration of the soil preventing considerable amounts of N_2O released from denitrification. Under such conditions, N_2O emissions were shown to be generally low (Pelster et al., 2012; Jamali et al., 2016). This might have been one reason for the comparably low FRE. Another reason for the low FRE might have been the mild winters without distinct frost/thaw cycles at all study sites.

With the new OSR specific emission factor of 0.6 % for the calculation of direct N_2O emissions from fertilizer N input GHG savings are possible up to 56 %. Herewith the stipulated criteria from the Renewable Energy Directive for biofuels can be fulfilled.

Can nitrification inhibitors help to reduce N_2O emission from OSR?

Ruser & Schulz (2015) updated the data set of the meta-analysis from *Akiyama* et al. (2010) with recent results from field studies published between 2010 – 2013. In total, they expanded the data from 85 to 140 sets measured on arable fields and grassland with different types of fertilizer and nitrification inhibitors (NIs). They reported a mean reduction of N_2O emission by approx. 35 % and concluded that NIs are a potent mitigation option for nitrous oxide emissions. Among the 140 data sets were only 28 annual data sets. The other data sets were not based on annual measurements or did not consider the winter period or they were determined under dry soil moisture conditions unrepresentative for German climate conditions. Therefore, the effect of NIs on winter nitrous oxide fluxes remained unclear. The

mitigation option of the nitrification inhibitor (NI) Piadin® combined with the organic fertilizer digestate was discussed in detail in Chapter 5. Across regions and years of the investigation, N_2O emissions were reduced by 36 % due to the use of NI. The reduction is corresponding with other studies reporting a reduction between 35 and 92 % (*Akiyama* et al., 2010; *Menendez* et al., 2012; *Ruser & Schulz*, 2015). However, with regard to annual N_2O emission data sets this mitigation effect was only three times significant indicating that environmental conditions may differently modify the mitigation potential between the single experimental years.

In the fertilization period, the mitigation effect became more obvious. In this period, the mean fluxes showed mitigation between 69 and 97 %, six of the 15 data sets were statistically significant. *Wolf* et al. (2014) reported a reduction of 62 % during the weeks after digestate fertilization, whereas they did not find any difference between their treatments on an annual base. This demonstrates the importance of annual datasets to prevent an overestimation of the effect of NIs on N_2O emission.

In Chapter 6 the mitigation potential of the nitrification inhibitor DMPP in combination with a mineral fertilizer at the site Hohenheim was discussed in detail. There was a significant mitigation effect (17 %) in the first year of the investigation when DMPP was applied together with ammonium nitrate sulphate. At the site Hohenheim, water-filled pore space was often below 60 %. Many studies reported a threshold value of > 60 % WFPS above which N_2O emissions strongly increased (*Dobbie* et al., 1999; *Skiba & Ball*, 2002; *Batemann & Baggs*, 2005). As we seldom reached this threshold value, low WFPS was probably the reason for the low fluxes connected with a low reduction potential of the NI.

In both cases (mineral and organic fertilizers) NIs are a promising tool to mitigate fertilizer driven N_2O emissions in OSR. However, this effect seems to be strongly depending on site, soil and weather conditions. It can therefore be assumed that the mitigation potential of NIs may not be fully exploited.

Do OSR crop residues contribute to N_2O release after harvest?

The effect of OSR residues on direct nitrous oxide emissions under field conditions was discussed in detail in Chapter 7 and 8. Approximately 0.4 million tons of global N_2O-N yr^{-1} production are emitted during the turnover of crop residues (*Moiser* et al., 1998). Several studies reported increased N_2O emissions after the incorporation of crop residues (*Flessa & Beese*, 1995; *Baggs* et al., 2000; *Millar & Baggs,* 2004; *Chen* et al., 2013). Crop residue induced emission are negative correlated with the C/N-ratio of the residues (*Kaiser* et al., 1998) and dependent on soil moisture, pH and soil texture (*Chen* et al., 2013). When compared to a control without crop residues, *Chen* et al. (2013) reported slightly higher N_2O emissions for treatments with C/N-ratios even between 45 and 100.

The crop residues in our field investigation had a C/N-ratio of 52, therefore increased nitrous oxide fluxes could also be expected. However, no significant difference between crop residue

amended and control treatment without crop residues was measured. Only 4.2 % (conventional tillage) and 5.2 % (reduced tillage) of the N released as N_2O during the investigated period came directly from the applied OSR crop residues. Nitrous oxide emissions from crop residue N occurred mainly in the first two months before tillage. This corresponds with *Baggs* et al. (2000) who found 65 % of the measured N_2O emissions during two weeks after the soil amendment with crop residues. This can be interpreted as rapid stimulation of microbial decomposition of the crop residues (*Shen* et al., 1989) related to anaerobic conditions resulting from microbial respiration which, together with increased C supply, as substrate for denitrification favored higher N_2O emissions. Hence, the low contribution from crop residues on total N_2O emissions in our investigation presumable resulted from the large C/N–ratio of the OSR crop residues. This conclusion was supported by the results of the microcosm trial in Chapter 8. In this investigation different amounts and qualities of OSR crop residues were tested on their effect on nitrous oxide emissions. There were no significant differences between the treatments with crop residues and the unamended treatments for cumulative N_2O emission. It can be assumed that the incorporated OSR crop residues led to a fast NO_3^- immobilization due to the high C/N values between 55 and 102 (*Powlson* et al., 2001; *Smith & Smith*, 2009), thus lowering substrate availability for denitrification. *Chaves* et al. (2007) observed a low share of plant available NO_3^--N in soil after incorporation of organic materials with high C/N–ratios. *Kaewpradit* et al. (2008) observed in this context a reduction of N_2O emissions. It is highly probable that also in our field and microcosm trials N was the limiting factor in treatments amended with crop residues. Consequently, the contribution of crop residues was only of marginal importance for the N_2O release after OSR harvest.
The general opinion of high N balances due to OSR crop residues was not confirmed in these experiments.

Does tillage affect N_2O emission from OSR production?

The effect of reduced tillage (RT) compared with conventional tillage (CT) on nitrous oxide emissions was discussed in detail in Chapter 7 and 9. There was no significant difference between the treatments in any of the investigated years. The main reasons for similar emissions from both tillage systems were probably the high temporal and spatial variability in N_2O emission data, which impeding a detection of statistically significant differences between the two tillage systems (*Kravchenko & Robertson*, 2015). Furthermore, Rochette (2008) reported that no-tillage systems promote N_2O emissions in poorly aerated soils, but with no impact in good and medium aerated soils, like the soil in this investigation. Similar N_2O emissions from RT and CT systems have also been reported by *Chatskikh* et al. (2008), *Liu* et al. (2006), *Abdalla* et al. (2010), and by *Negassa* et al. (2015).
But results from tillage investigations are contractionary, there are several studies which reported higher N_2O emissions under RT compared to CT associated with increased rates of

denitrification, greater bulk density and soil moisture (*Gossens* et al., 2001; *D'Haene* et al., 2008; *Abdalla* et al., 2013; *Lognoul et* al., 2017). However, there are studies who reported lower N_2O emission from RT systems due to the lower mixing of fertilizer and crop residues into the soil (*Chatskikh* & *Olesen* 2007; *Mutegi* et al., 2010; *Koga*, 2013).

Conclusions

The study has shown that the common amount of N-fertilizer with 180 kg N ha^{-1} does not result in higher oil yield compared to a fertilization with 120 kg N ha^{-1}. Thus, direct as well as indirect N_2O emission can be reduced under a lower N-fertilization rate. Overall, N_2O emission was comparatively low in all treatments and in all years of this investigation. However, mitigation strategies could be demonstrated due to the reduction of N-fertilizer and nitrification inhibitors. Contrary to the original assumptions, OSR crop residues had no promoting effect on the N_2O emission.

11 References

Abbasi MK, Adams WA (2000): Gaseous N emission during simultaneous nitrification–denitrification associated with mineral N fertilization to a grassland soil under field conditions. Soil Biol Biochem 32, 1251–1259.

Abdalla M, Jones M, Ambus P, Williams M (2010): Emissions of nitrous oxide from Irish arable soils: effects of tillage and reduced N input. Nutr Cycl Agroecosys 86, 53–65.

Abdalla M, Osborne B, Lanigan G, Forristal D, Williams M, Smith P, Jones MB (2013): Conservation tillage systems: a review of its consequences for greenhouse gas emissions. Soil Use Manage 29, 199–209.

Akiyama H, Tsuruta H, Watanabe T (2000): N_2O and NO emissions from soils after the application of different chemical fertilizers. Chemosphere 2, 313–320.

Akiyama H, Yan X, Yagi K (2010): Evaluation of effectiveness of enhanced-efficiency fertilizers as mitigation options for N_2O and NO emissions from agricultural soils: meta-analysis. Glob Change Biol 16, 1837–1846.

Alonso–Ayuso M, Gabriel JL, Quemada M (2016): Nitrogen use efficiency and residual effect of fertilizers with nitrification inhibitors. Eur J Agron 80, 1–8.

Alvarez R (2005): A review of nitrogen fertilizer and conservation tillage effects on soil organic carbon storage. Soil Use Manage 21, 38–52.

Arah JRM, Smith KA, Crichton IJ, Li HS (1991): Nitrous oxide production and denitrification in Scottish arable soils. J Soil Sci 42, 351–367.

Arp DJ, Stein LY (2003): Metabolism of inorganic N compounds by ammonia-oxidizing bacteria. Crit Rev Biochem Mol Bio 38, 471–495.

Arregui LM, Quemada M (2008): Strategies to improve nitrogen use efficiency in winter cereal crops under rain fed conditions. Agron J 100, 277–284.

Aulakh MS, Khera TS, Doran JW, Bronson KF (2001): Denitrification, N_2O and CO_2 fluxes in rice-wheat cropping system as affected by crop residues, fertilizer N and legume green manure. Biol Fertil Soils 34, 375–389.

Azam F, Müller C, Weiske A, Benckiser G and Ottow JCG (2002): Nitrification and denitrification as sources of atmospheric nitrous oxide – role of oxidizable carbon and applied nitrogen. Biol Fertil Soils 35, 54–61.

Baggs EM (2011): Soil microbial sources of nitrous oxide: recent advances in knowledge, emerging challenges and future direction. Curr Opin Environ Sustain 3, 321–327.

Baggs EM, Rees R, Smith KA, Vinten AJA (2000): Nitrous oxide emission from soils after incorporating crop residues. Soil Use Manage 16, 82–87.

Baggs EM, Stevenson M, Pihlatie M, Regar A, Cook H, Cadisch G (2003): Nitrous oxide emissions following application of residues and fertilizer under zero and conventional tillage. Plant Soil 254, 361–370.

Ball BC, Crichton I, Horgan GW (2008): Dynamics of upward and downward N_2O and CO_2 fluxes in ploughed or no-tilled soils in relation to water-filled pore space, compaction and crop presence. Soil Tillage Res 101, 20–30.

Ball BC, Scott A, Parker JP (1999): Field N_2O, CO_2 and CH_4 fluxes in relation to tillage, compaction and soil quality in Scotland. Soil Till Res 53, 29–39.

Bartling J, Kotzerke A, Mai M, Esperschütz J, Buegger F, Schloter M, Wilke B M (2009): Microbial community structure and function during abnormal curve development of substrate-induced respiration measurements. Chemosphere 77, 1488–1494.

Bateman EJ, Baggs EM (2005): Contributions of nitrification and denitrification to N_2O emissions from soils at different water-filled pore space. Biol Fertil Soil 41, 379–388.

Beauchamp EG, Trevors JT, Paul JW (1989): Carbon sources for bacterial denitrification. Adv Soil Sci 10, 113–142.

Bouwman AF (1996): Direct emission of nitrous oxide from agricultural soils. Nutr Cycl Agroecosyst 46, 53–70.

Bouwman AF, Boumans LJM, Batjes NH (2002): Emissions of N_2O and NO from fertilized fields: summary of available measurement data. Glob Biogeochem Cycles 16, 1058.

Bremner JM (1997): Sources of nitrous oxide in soils. Nutr Cycl Agroecosyst 49, 7–16.

Butterbach-Bahl K, Baggs EM, Dannenmann M, Kiese R, Zechmeister-Boltenstern S (2013): Nitrous oxide emissions from soils. How well do we understand the processes and their controls? Philos T Roy Soc B 368 (1621), 20130122.

Carré P, Pouzet A (2014): Rapeseed market, worldwide and in Europe. OCL 21, D102.

Chatskikh D, Olesen JE (2007): Soil tillage enhanced CO_2 and N_2O emissions from loamy sand soil under spring barley. Soil Till Rese 97, 5–18.

Chatskikh D, Olesen JE, Hansen EM, Elsgaard L, Petersen B M (2008): Effects of reduced tillage on net greenhouse gas fluxes from loamy sand soil under winter crops in Denmark. Agric Ecosyst Environ 128, 117–126.

Chaves B, De Neve S, Boeckx P, Van Cleemput O, Hofman G (2007): Manipulating nitrogen release from nitrogen-rich crop residues using organic wastes under field conditions. Soil Sci Soc Am J 71, 1240–1250.

Chen H, Li X, Hu F, Shi W (2013): Review: Soil nitrous oxide emissions following crop residue addition: a meta-analysis. Glob Change Biol 19, 2956–2964.

Clais P, Sabine C, Bala G, Bopp L, Brovkin V, Canadell J, Chhabra A, DeFries R, Galloway J, Heimann M, Jones C, Le Quéré C, Myneni RB, Piao S and Thornton P (2013): Carbon and Other Biogeochemical Cycles. In: Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change [Stocker, T.F., D. Qin, G.-K. Plattner, M. Tignor, S.K. Allen, J. Boschung, A. Nauels, Y. Xia, V. Bex and P.M. Midgley (eds.)]. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

Copec K, Filipovic D, Husnjak S, Kovacev I, Kosutic S (2015): Effects of tillage systems on soil water content and yield in maize and winter wheat production. Plant Soil Environ 61, 213–219.

Crutzen PJ (1981): Atmospheric chemical processes of the oxides of nitrogen including nitrous oxide. In: Delwiche, C.C. (Ed.), Denitrification, Nitrification and Atmospheric N_2O. Wiley, Chichester, 17–44.

Cunningham MH, Chaney K, Bradbury RB and Wilcox A (2004): Non-inversion tillage and farmland birds: a review with special reference to UK and Europe. Ibis, 146 (Suppl. 2), 192–202.

D′ Haene K, Van Den Bossche A, Vandenbruwane J, De Neve S, Gabriels D, Hofman G (2008): The effect of reduced tillage on nitrous oxide emissions of silt loam soils. Biol Fertil Soils 45, 213–217.

Davidson EA (1992): Sources of nitric oxide and nitrous oxide following wetting of dry soil. Soil Sci Soc Am J 56, 95–102.

Di HJ and Cameron KC (2007): Nitrate leaching losses and pasture yields as affected by different rates of animal urine nitrogen returns and application of a nitrification inhibitor–a lysimeter study. Nutr Cycl Agroecosyst 79, 281–290.

Dobbie KE, McTaggart IP, Smith KA (1999): Nitrous oxide emissions from intensive agricultural systems: variations between crops and seasons, key driving variables, and mean emission factors. J Geophys Res 21, 26891–26899.

Dobbie KE, Smith KA (2001): Impact of different forms of N fertilizer on N_2O emissions from intensive grassland. Nutr Cycl Agroecosyst 67, 37–46.

Edwards R, Mulligan D, Giuntoli J, Agostini A, Boulamanti A, Koeble R, Marelli L, et al. (2013): Assessing GHG Default Emissions from Biofuels in EU Legislation. Review of Input Database to Calculate Default GHG Emissions, Following Expert Consultation 22–23 November 2011, Ispra (Italy). JRC Scientific and Policy Reports. EUR 25595 EN. European Commission Joint Research Centre Institute for Energy and Transport.

Ercoli L, Masoni A, Pampana S, Mariotti M, Arduini I (2013): As durum wheat productivity is affected by nitrogen fertilisation management in Central Italy. Eur J Agron 44, 38–45.

EUROSTAST (2017): http://ec.europa.eu/eurostat/de/data/database. Access: 21.06.17.

Fangueiro D, Fernandes A, Coutinho J, Moreira N, Trindade H (2009): Influence of two nitrification inhibitors (DCD and DMPP) on annual ryegrass yield and soil mineral N dynamics after incorporation with cattle slurry. Commun Soil Sci Plant Anal 40, 3387–3398.

FAOSTAT (2017): http://www.fao.org/faostat/en/#data. Access: 21.06.2017.

Firestone MK and Davidson EA (1989): Microbiological basis of NO and N_2O production and consumption in soil. In: Exchange of Trace Gases Between Terrestrial Ecosystems and the Atmosphere, edited by: Andreae MO & Schimel DS, Wiley, 721.

Flessa H and Beese F (1995): Effects of Sugarbeet Residues on Soil Redox Potential and Nitrous Oxide Emission. Soil Sci Soc Am J 59, 1044–1051.

Flessa H, Dorsch P and Beese F (1995): Seasonal variation of N_2O and CH_4 fluxes in differently managed arable soils in Southern Germany. J Geophys Res 100, 23115–23124.

Flessa H, Wild U, Klemisch M, Pfadenhauer J (1998): Nitrous oxide and methane fluxes from organic soils under agriculture. Eur J Soil Sci 49, 327–335.

FNR (2017): https://www.fnr.de/nachwachsende-rohstoffe/bioenergie/biokraftstoffe. Access: 21.06.17.

Fuß R, Ruth B, Schilling R, Scherb H, Munch JC (2011): Pulse emissions of N_2O and CO_2 from an arable field depending on fertilization and tillage practice. Agri Ecosys Environ 144, 61–68.

Garcia-Montiel DC, Melillo JM, Steudler PA, Cerri CC, Piccolo MC (2003): Carbon limitations to nitrous oxide emissions in a humid tropical forest of the Brazilian amazon. Biol Fertil Soils 38, 267–272.

Garcia-Ruiz R, Baggs EM (2007): N_2O emission from soil following combined application of fertiliser-N and ground weed residues. Plant Soil 299, 263–274.

Gillam KM, Zebarth BJ, Burton DL (2008): Nitrous oxide emissions from denitrification and the partitioning of gaseous losses as affected by nitrate and carbon addition and soil aeration. Can J Soil Sci 88, 133–143.

Goossens A, Visscher A, Boeckx P, Van Cleemput O (2001): Two year field study on the emission of N_2O form coarse and middle-textured Belgian soils with different land use. Nutr Cycl Agroecosyst 60, 23–34.

Goreau TJ, Kaplan WA, Wofsy SC, McElroy MB, Valois FW, Watson SW (1980): Production of NO^- and N_2O by nitrifying bacteria at reduced concentrations of oxygen. Appl Environ Microbiol 40, 526–532.

Goyal S, Mishra MM, Hooda IS, Singh R (1992): Organic matter-microbial biomass relationships in field experiments under tropical conditions: Effects of inorganic fertilization and organic amendments. Soil Biol Biochem 24, 1081–1084.

Granli T, Bøckman OC (1994): Nitrous oxide from Agriculture. Norwegian J Agri Res Supplement 12.

Gregorich EG, Kachanoski RG, Voroney RP (1989): Carbon mineralization in soil size fractions after various amounts of aggregate disruption. J Soil Sci 40, 649–659.

Gregorich EG, Rochette P, Hopkins DW, McKim UF, StGeorges P (2006): Tillage-induced environmental conditions in soil and substrate limitation determine biogenic gas production. Soil Biol Biochem 38, 2614–2628.

Gruber S, Pekrun C, Möhring J, Claupein W (2012): Long-term yield and weed response to conservation and stubble tillage in SW Germany. Soil Till Res 121, 49–56.

Halvorson AD, Del Grosso SJ, Alluvione F (2010): Nitrogen source effect on nitrous oxide emissions from irrigated no-till corn. J Environ Qual 39, 1554–1562.

Hamelinck C, De Loveinfosse I, Koper M et al. (2012): Renewable Energy Progress and Biofuels Sustainability Ecofys, London, 450.

Hayatsu M, Tago K, Saito M (2008): Various players in the nitrogen cycle: Diversity and functions of the microorganisms involved in nitrification and denitrification. Soil Sci Plant Nutr 54, 33-45.

Hénault C, Grossel A, Mary B, Roussel M, Léonard J (2012): Nitrous oxide emission by agricultural soils: a review of spatial and temporal variability for mitigation. Pedosphere 22, 426–433.

Henke J, Breustedt G, Sieling K, Kage H (2007): Impact of uncertainty on the optimum nitrogen fertilization rate and agronomic, ecological and economic factors in an oilseed rape based crop rotation. J Agric Sci 145, 455-468.

Herr C (2015): Einfluss von Rapsernterückständen auf umweltrelevante Nährstoffausträge in einem Mikrokosmenversuch. Master thesis, University of Hohenheim, Germany.

Hoben JP, Gehl RJ, Millar N, Grace PR, Robertson GP (2011): Nonlinear nitrous oxide (N_2O) response to nitrogen fertilizer in on-farm corn crops of the US Midwest. Gobal Change Biol 17, 1140-1152.

Hoefnagels R, Smeets E, Faaij A (2010): Greenhouse gas footprints of different biofuel production systems. Renew Sust Energ Rev 14, 1661–1694.

Holland JM (2004): The environmental consequences of adopting conservation tillage in Europe: reviewing the evidence. Agric Ecosyst Environ 103, 1–25.

i.m.a. (2005): Landwirtschaft in Deutschland. i.m.a – information.medien.agrar e.V., Bonn.

IPCC (2006): Intergovernmental Panel on Climate Change, 2006. Guidelines for National Greenhouse Gas Inventories. Geneva, Switzerland. Available at http://www.ipcc-nggip.iges.or.jp/public/2006gl/index.html. Access 21.06.17.

IPCC (2013): Summary for Policymakers. In: Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change [Stocker TF, Qin D, Plattner GK, Tignor M, Allen SK, Boschung J, Nauels A, Xia Y, Bex V and Midgley PM (eds.)]. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

Jamali H, Quale W, Scheer C, Rowlings D, Baldock J (2016): Effect of soil texture and wheat plants on N_2O fluxes: A lysimeter study. Agric Forest Meteorol 223, 17–29.

Johnson J, Reicosky D, Allmaras R, Sauer T, Venterea R, Dell C (2005): Greenhouse gas emissions and mitigation potential of agriculture in central USA. Soil Till Res 83, 73–94.

Jungkunst HF, Freibauer A, Neufeldt H, Bareth G (2006): Nitrous oxide emissions from agricultural land use in Germany – a synthesis of available annual field data. J Plant Nutr Soil Sci 169, 341–351.

Kaewpradit W, Toomsan B, Vityakon P, Limpinuntana V, Saenjan P, Jogloy S, Patanothai A, Cadisch G (2008): Regulating mineral N release and greenhouse gas emissions by mixing groundnut residues and rice straw under field conditions. Eur J Soil Sci 59, 640–652.

Kaiser EA, Kohrs K, Kücke M, Schnug E, Heinemeyer O, Munch JC (1998): Nitrous oxide release from arable soil: importance of N-fertilisation, crops and temporal variation. Soil Biol Biochem 30, 1553–1563.

Kaiser EA, Kohrs K, Kücke M, Schnug E, Heinemeyer O, Munch JC (1998): Nitrous oxide release from arable soil: importance of N-fertilisation, crops and temporal variation. Soil Biol Biochem 30, 1553–1563.

Kaiser EA, Ruser R (2000): Nitrous oxide emissions from arable soils in Germany – An evaluation of six long-term field experiments. J. Plant Nutr Soil Sci 163, 249–260.

Khalil M and Baggs EM (2005): CH_4 oxidation and N_2O emissions at varied soil water-filled pore spaces and headspace CH_4 concentrations. Soil Biol Biochem 37, 1785–1794.

Kim DG, Hernandez-Ramirez G, Giltrap D (2013): Linear and nonlinear dependency of direct nitrous oxide emissions on fertilizer nitrogen input. A meta-analysis. Agric Ecosys Environ 168, 53–65.

Knowles R (1982): Denitrification. Microbiol Rev 46, 43–70.

Koga N (2013): Nitrous oxide emissions under a four-year crop rotation system in northern Japan: impacts of reduced tillage, composted cattle manure application and increased plant residue input. Soil Sci Plant Nutr 59, 56–68.

Kool DM, Dolfing J, Wrage N and van Groenigen JW (2011): Nitrifier denitrification as a distinct and significant source of nitrous oxide from soil. Soil Biol Biochem 43, 174–178.

Kravchenko AN and Robertson GP (2015): Statistical challenges in analyses of chamber-based soil CO_2 and N_2O emissions data. Soil Sci Soc Am J 79, 200–211.

Ladha JK, Pathak H, Krupnik TJ, Six J, van Kessel C (2005): Efficiency of fertilizer nitrogen in cereal production: retrospects and prospects. Adv Agron 87, 85–156.

Lal R (2005): World crop residues production and implications of its use as a biofuel. Environ Int 31, 575–584.

Lampurlanés J, Angas P, Cantero-Martinez C (2001): Root growth: soil water content and yield of barley under different tillage systems on two soils in semiarid conditions. Field Crops Res 69, 27–40.

Lehtinen T, Schlatter N, Baumgarten A, Bechini L, Krüger J, Grignani C, Zavattaro L, Costamagna C, Spiegel H (2014): Effect of crop residue incorporation on soil organic carbon and greenhouse gas emissions in European agricultural soils. Soil Use Manage 30, 524–538.

Li H, Liang X, Chen Y, Lian Y, Tian G, Ni W (2008): Effect of nitrification inhibitor DMPP on nitrogen leaching, nitrifying organisms, and enzyme activities in a rice-oilseed rape cropping system. J Environ Sci 20(2)149–155.

Li X, Sørensen P, Olesen JE, Petersen SO (2016): Evidence for denitrification as main source of N_2O emission from residue-amended soil. Soil Biol Biochem 92, 153–160.

Linn DM, Doran JW (1984): Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and nontilled soils. Soil Sci Soc Am J 48, 1267–1272.

Liu C, Wang K, Zheng X (2013): Effects of nitrification inhibitors (DCD and DMPP) on nitrous oxide emission: crop yield and nitrogen uptake in a wheat-maize cropping system. Biogeosciences 10, 2427–2437.

Liu XJ, Mosier AR, Halvorson AD & Zhang FS (2006): The impact of nitrogen placement on NO, N_2O, CH_4 and CO_2 fluxes from non-tilled soil compared with conventional-tilled soil. Plant Soil 280, 177–188.

Lognoul M, Theodorakopoulos N, Hiel M, Regaert D, Broux F, Heinesch B et al. (2017): Impact of tillage on greenhouse gas emissions by an agricultural crop and dynamics of N_2O fluxes. Insights from automated closed chamber measurements. Soil Till Res 167, 80–89.

MacKenzie AF, Fan MX, Cadrin F (1997): Nitrous oxide emission as affected by tillage, corn-soybean-alfalfa and nitrogen fertilization. Can J Soil Sci 77, 145-152.

Malhi SS and Lemke R (2007): Tillage, crop residue and N fertilizer effects on crop yield, nutrient uptake, soil quality and nitrous oxide gas emissions in a second 4-yr rotation cycle. Soil Till Res 96, 269–283.

Martinez F, Palencia P, Weiland CM, Alonso D, Oliveira JA (2015): Influence of nitrification inhibitor DMPP on yield: fruit quality and SPAD values of strawberry plants. Sci Hortic 185, 233–239.

Menendez S, Barrena I, Setien I, Gonzalez-Murua C, Estavillo JM (2012): Efficiency of nitrification inhibitor DMPP to reduce nitrous oxide emissions under different temperature and moisture conditions. Soil Biol Biochem 53, 82–89.

Millar N, Baggs EM (2004): Chemical composition, or quality, of agroforestry residues influences N_2O emissions after their addition to soil. Soil Biol Biochem 36, 935–943.

Miller N, Zebarth BJ, Dandie CE, Burton DL, Goyer C, Trevors JT (2008): Crop residue influence on denitrification, N_2O emissions and denitrifier community abundance in soil. Soil Biol Biochem 40, 2553–2562.

Möller K and Müller T (2012): Effects of anaerobic digestion on digestate nutrient availability and crop growth. A review. Eng Life Sci 12, 242–257.

Mosier A, Kroeze C, Nevison C, Oenema O, Seitzinger S and van Cleemput O (1998): Closing the global N_2O budget: nitrous oxide emissions through the agricultural nitrogen cycle – OECD/IPCC/IEA phase II development of IPCC guidelines for national greenhouse gas inventory methodology. Nutr Cycl Agroecosys 52, 225–248.

Mosier AR, Parton WJ, Hutchinson GL (1983): Modelling nitrous oxide evolution from cropped and native soils. In: Hallberg, R. (Ed.), Environmental Biogeochemistry. Ecological Bulletin, Stockholm, 229–241.

Mutegi JK, Munkholm LJ, Petersen BM, Hansen EM, Petersen SO (2010): Nitrous oxide emissions and controls as influenced by tillage and crop residue management strategy. Soil Biol Biochem 42, 1701–1711.

Myhre G, Shindell D, Bréon FM, Collins W, Fuglestvedt J, Huang J, Koch D, Lamarque JF, Lee D, Mendoza B, Nakajima T, Robock A, Stephens G, Takemura T and Zhang H (2013): Anthropogenic and Natural Radiative Forcing. In: Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change [Stocker TF, Qin D, Plattner GK, Tignor M. Allen SK, Boschung J, Nauels A, Xia Y, Bex V and Midgley PM (eds.)]. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

Negassa W, Price RF, Basir A, Snapp SS, Kravchenko A (2015): Cover crop and tillage systems effect on soil CO_2 and N_2O fluxes in contrasting topographic positions. Soil Tillage Res 154, 64–74.

Oades JM (1984): Soil organic matter and structural stability: mechanisms and implications for management. Plant Soil 76, 319–337.

Palma RM, Rimolo M, Saubidet MI, Conti ME (1997): Influence of tillage system on denitrification in maize-cropped soils. Biol Fert Soil 25, 142–146.

Paré MC, Lafond J, Pageau D (2015): Best management practices in Northern agriculture. A twelve-year rotation and soil tillage study in Saguenay–Lac-Saint-Jean. Soil Till Rese 150, 83–92.

Parkin TB and Robinson JA (1993): Statistical evaluation of median estimators for lognormally distributed variables. Soil Sci Soc Am J 57, 317–323.

Pasda G, Hahndel R, Zerulla W (2001): Effect of fertilizers with the new nitrification inhibitor DMPP (3,4-dimethylpyrazole phosphate) on yield and quality of agricultural and horticultural crops. Biol Fert Soil 34, 85–97.

Pelster DE, Chatigny MH, Rochette P, Angers DA, Rieux C, Vanasse A (2012): Nitrous oxide emissions respond differently to mineral and organic nitrogen sources in contrasting soil types. J Environ Qual 41, 427–435.

Pfab H (2011): Nitrous oxide emissions and mitigation strategies: measurements on an intensively fertilized vegetable cropped loamy soil. PhD thesis, University of Hohenheim, Germany.

Pfab H, Palmer I, Buegger F, Fiedler S, Müller T, Ruser R (2012): Influence of a nitrification inhibitor and of placed N-fertilization on N_2O fluxes from a vegetable cropped loamy soil. Agric Ecosyst Environ 150, 91–101.

Powlson DS, Hirsch PR, Brookes PC (2001): The role of soil microorganisms in soil organic matter conservation in the tropics. Nutr Cycl Agroecosys 61, 41–51.

Rathke GW, Behrens T, Diepenbrock W (2006): Integrated nitrogen management strategies to improve seed yield, oil content and nitrogen efficiency of winter oilseed rape (*Brassica napus* L.): A review. Agric Ecosys Environ 117, 80–108.

RED (2009): 28/EC of the European Parliament and of the Council of 23 April 2009 on the promotion of the use of energy from renewable sources and amending and subsequently repealing Directives 2001/77/EC and 2003/30/EC. Official Journal of the European Union 140, 16–45.

Robertson GP, Groffman PM (2007): Nitrogen transformations. In: Paul, E.A. (Ed.), Soil Microbiology, Chemistry, and Ecology. Springer, New York USA, pp. 341–364.

Rochette P (2008): No-till only increases N_2O emissions in poorly-aerated soils. Soil Till Res 101, 97–100.

Rochette P, Angers DA, Chantigny MH, Bertrand N (2008): N_2O emissions respond differently to no-till in a loam and a heavy clay soil. Soil Sci Soc Am J 72, 1363–1369.

Röver M, Heinemeyer O, Kaiser EA (1998): Microbial induced nitrous oxide emissions from an arable soil during winter. Soil Biol Biochem 14, 1859–1865.

Ruser R, Flessa H, Russow R, Schmidt G, Buegger F, Munch JC (2006): Emission of N_2O, N_2 and CO_2 from soil fertilized with nitrate. Effect of compaction, soil moisture and rewetting. Soil Biol Biochem 38, 263–274.

Ruser R, Flessa H, Schilling R, Beese F, Munch J C (2001): Effect of crop-specific field management and N fertilization on N_2O emissions from a fine-loamy soil. Nutr Cycl Agroecosys 59, 177–191.

Ruser R, Schilling R, Steindl H, Flessa H, Beese F (1998): Soil Compaction and Fertilization Effects on Nitrous Oxide and Methane Fluxes in Potato Fields. Soil Sci Soc Am J 62, 1587–1595.

Ruser R, Schulz R (2015): The effect of nitrification inhibitors on the nitrous oxide (N_2O) release from agricultural soils–a review. J Plant Nutr Soil Sc 178, 171–188.

Sanz-Cobena A, Sánchez-Martín L, García-Torres L, Vallejo A (2012): Gaseous emissions of N_2O and NO and NO_3^- leaching from urea applied with urease and nitrification inhibitors to a maize (Zea mays) crop. Agri Ecosys Environ 149, 64–73.

Shan J, Yan X (2013): Effects of crop residue returning on nitrous oxide emissions in agricultural soils. Atmos Environ 71, 170–175.

Shaw LJ, Nicol GW, Smith Z, Fear J, Prosser JI, Baggs EM (2006): Nitrosospira spp. can produce nitrous oxide via a nitrifier denitrification pathway. Environ Microbiol 8, 214–222.

Shcherbak I, Millar N, Robertson GP (2014): Global metaanalysis of the nonlinear response of soil nitrous oxide (N_2O) emissions to fertilizer nitrogen. PNAS 111, 9199–9204.

Shen SM, Hart PBS, Powlson DS (1989): The nitrogen cycle in the broadbalk wheat experiments N -labelled fertilizer residues in the soil and in the soil microbial biomass. Soil Biol Biochem 21, 529–533.

Sieling K and Kage H (2010): Efficient N management using winter oilseed rape. A review. Agron Sustain Dev 30, 271–279.

Skiba U, Ball B (2002): The effect of soil texture and soil drainage on emissions of nitric oxide and nitrous oxide. Soil Use Manage 18, 56–60.

Smith KA, McTaggart IP, Dobbie KE & Conen F (1998): Emissions of N_2O from Scottish agricultural soils, as a function of fertilizer N. Nutr Cycl Agroecosyst 52, 123–130.

Smith TM, Smith RL (2009): Ökologie (6th ed., p. 608). München: Pearson Education Deutschland GmbH.

Sölter K, Weber N (2000): Die Bedeutung der Säurekapazität auf Kläranlagen; Anwendungsbericht Ch No. 77 (p. 6). Düsseldorf.

Stange CF, Spott O, Arriaga H, Menendez S, Estavillo JM, Merino P (2013): Use of the inverse abundance approach to identify the sources of NO and N2O release from Spanish forest soils under oxic and hypoxic conditions. Soil Biol Biochemi 57, 45–1458.

STATISTA (2017): https://de.statista.com/statistik/daten. Access: 21.06.17.

Stehfest E, Bouwman A L (2006): N_2O and NO emission from agricultural fields and soils under natural vegetation: summarizing available measurement data and modeling of global annual emissions. Nutr Cycl Agroecosys 74, 207–228.

Sutton AL, Huber DM, Jones DD, Kelly DT, Bache DH (1986) Use of nitrification inhibitors and ammonia enrichment with swine manure applications. Appl Eng Agric 2, 179–185.

Tiedje JM (1988): Ecology of denitrification and dissimilatory nitrate reduction to ammonium. p. 179–244. In A.J.B. Zehnder (ed), Environmental Microbiology of Anaerobes. John Wiley and Sons, N.Y.

Van Groenigen JW, Velthof GL, Oenema O, van Groenigen K J, van Kessel C (2010): Towards an agronomic assessment of N_2O emissions. A case study for arable crops. Eur J Soil Sci 61, 903–913.

Velthoff GL, Kuikman PJ, Oenema O (2002): Nitrous oxide emission from soils amended with crop residues. Nutr Cycl Agroecosys 55, 757–761.

Venterea R, Burger M and Spokas K (2005): Nitrogen Oxide and Methane Emissions under Varying Tillage and Fertilizer Management. J Environ Qual 34, 1467–1477.

Venterea RT, Bijesh M, Dolan MS (2011): Fertilizer source and tillage effects on yield-scaled nitrous oxide emissions in a corn cropping system. J Environ Qual 40, 1521–1531.

Walter K, Don A, Fuß R, Kern J, Drewer J, Flessa H (2015): Direct nitrous oxide emissions from oilseed rape cropping - a meta-analysis. Glob Change Biol Bioenergy 7, 1260–1271.

Wang W, Dalal R (2015): Nitrogen management is the key for low-emission wheat production in Australia: a life cycle perspective. Eur J Agron 66, 74–82.

Weber DF, Gainey PL (1962): Relative sensitivity of nitrifying organisms to hydrogen ions in soils and solutions. Soil Sci 94, 138–145.

Weiske A, Benckiser G, Herbert T, Ottow J (2001): Influence of the nitrification inhibitor 3,4-dimethylpyrazole phosphate (DMPP) in comparison to dicyandiamide (DCD) on nitrous oxide emissions, carbon dioxide fluxes and methane oxidation during 3 years of repeated application in field experiments. Biol Fertil Soils 34, 109–117.

Weiske A, Benckiser G, Herbert T, Ottow JCG (2001): Influence of the nitrification inhibitor 3,4-dimethylpyrazole phosphate (DMPP) in comparison to dicyandiamide (DCD) on nitrous oxide emissions, carbon dioxide fluxes and methane oxidation during 3 years of repeated application in field experiments. Biol Fert Soils 34, 109–117.

WMO (2016): WMO Greenhouse Gas Bulletin No. 12.

Wolf U, Fuß R, Höppner F, Flessa H (2014): Contribution of N_2O and NH_3 to total greenhouse gas emission from fertilization. Results from a sandy soil fertilized with nitrate and biogas digestate with and without nitrification inhibitor. Nutr Cycl Agroecosyst 100, 121–134.

Wrage N, Velthof GL, Laanbroek HJ, Oenema O (2004): Nitrous oxide production in grassland soils: assessing the contribution of nitrifier denitrification. Soil Biol Biochem 36, 229–236.

Wrage N, Velthof GL, van Beusichem ML, Oenema O (2001): Role of nitrifier denitrification in the production of nitrous oxide. Soil Biol Biochem 33, 1723–1732.

Wu D, Senbayram M, Well R, Brüggemann N, Pfeiffer B, Loick N et al. (2017): Nitrification inhibitors mitigate N_2O emissions more effectively under straw-induced conditions favouring denitrification. Soil Biol Biochem 104, 197–207.

Yamamoto A, Akiyama H, Nakajima Y, Hoshino YT (2017): Estimate of bacterial and fungal N_2O production processes after crop residue input and fertilizer application to an agricultural field by ^{15}N isotopomer analysis. Soil Biol Biochem 108, 9–16.

Zerulla W, Barth T, Dressel J, Erhardt K, von Locquenghien KH, Pasda G, Rädle M, Wissemeier A (2001a): 3,4-dimethylpyrazole phosphate (DMPP)—A new nitrification inhibitor for agriculture and horticulture. Biol Fertil Soils 34, 79–84.

Zerulla W, Pasda G, Hähndel R, Wissemeier A (2001b): The new nitrification inhibitor DMPP (ENTEC®) for use in agricultural and horticultural crops - An overview. Plant Nutr 92, 754–755.

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